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ANALYSIS

OF

MILK AND MILK PRODUCTS

BY

HENRY LEFFMANN, M.A., M.D.,

AND

WILLIAM BEAM, M.A., M.D.

SECOND EDITION, REVISED AND ENLARGED,
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PREFACE.

The first edition of this book was quite favorably received by competent critics, but it was noted that the analysis of butter and cheese was too briefly treated. It is hoped that this objection has been removed in the present edition. A large amount of matter has been added concerning those topics; all the processes of analysis which have practical value in detecting butter adulteration have been given in detail, many from the latest publication of the A. O. A. C. A large addition has been made to the Chapter on Cheese, and a brief note on the fermented milk beverages. Other features have not been neglected. Considerable matter has been added in relation to the determination of lactose, among which may be mentioned Wiley and Ewell's recently described process of double dilution and polarization, and Allen's manipulation of Pavy's solution. The tables for determining total solids by Hehner and Richmond's formula have been recalculated according to Richmond's corrected formula.

Concerning the rapid method of fat-extraction with which our names are connected, we desire to say that the taking out of a patent was rendered necessary by some business arrangements over which we had no control and which, at that time, seemed to require such action to protect a reputable merchant from serious loss. The amount

paid was small and an offer has since been made to repurchase the patent for the sum originally paid for it, the intention being to make its use free, but the negotiations failed. We also deem it proper to say that the originality of the discovery rests only on the chemicals employed in liberating the fat. The application of centrifugal machinery to the separation of liquids of different specific gravity dates from more than a generation ago and cannot be now allowed as an invention. We have regretted that a foreign chemist has appropriated, without acknowledgment, the special features of our method.

As in the first edition, we have relied largely upon the contributions published in the "Proceedings of the Association of Official Agricultural Chemists," and in *The Analyst.*

H. L.
W. B.

715 Walnut Street, Philadelphia,
September, 1896.

CONTENTS.

NATURE AND COMPOSITION OF MILK.

Milk, the nutritive secretion of nursing mammals, consists of water, fat, proteids, sugar, and mineral matters. Cow's milk, being of the greatest importance to the analyst, will receive the largest share of attention, and will be understood to be meant in all cases, unless otherwise stated.

With rare exceptions, the secretion of milk takes place only as a result of pregnancy and delivery at term, and continues for a variable period. The chemistry of its formation is not entirely understood. The organic ingredients do not exist in appreciable quantities in the blood, and must, therefore, be elaborated by specific secretory action.

Fat.—This occurs in globules varying from .0015 mm. to .005 mm. in diameter, in a condition which prevents spontaneous coalescence. According to Gutzeit, the average size of the globules is affected by many circumstances, such as change of weather, of food, or locality. During the whole time of lactation they regularly diminish in diameter. Among cows of the same breed their average size, extending over the whole lactiferous period, is fairly constant. The properties exhibited by the fat-globules have been regarded as indicating the existence of a membrane surrounding them, but they may be explained without such an assumption. If an envelope of any kind exists, it probably is merely a liquid one, such as the film of a soap-bubble.

The fat of milk consists of a mixture of the ethers of tritenyl (C_3H_5), but is peculiar among animal fats in containing a notable proportion of acid-radicles with a small number of carbon atoms. Thus, about 91 per cent. consists of stearin, palmitin, and olein, and the remaining nine per cent. of butyrin and caproin along with minute amounts of caprin, myristin and caprylin, and some others. The exact arrangement of these constituents is not known, but the weight of opinion is that milk-fat is not a mixture of simple fats, but that several acid radicles are united to the same tritenyl molecule.

The following results of the analysis of a sample of butter-fat are given by Dr. Bell:—

	Per cent.
Butyric acid,	6.1
Caproic, caprylic, and capric acids,	2.1
Myristic, palmitic, and stearic acids,	49.4
Oleic acid,	36.1
Glycerol (calculated),	12.5

According to Duclaux, the mean composition of butter-fat is probably:—

	Per cent.
Palmitin, stearin, olein, with traces of myristin and butin,	91.5
Butyrin,	4.2
Capronin,	2.5
Caprylin, caprinin, laurin (traces),	1.8
	100.0

Proteids.—The nature of the proteids of milk has been much discussed, but it is now generally conceded that there are at least three forms, casein, albumin, and globulin, the casein being present in by far the greater amount, and the globulin as traces only.

Casein.—Casein is in large part, at least, in a gelatinous

(colloidal) form, probably in combination with calcium phosphate. It is precipitated from this condition by acids, rennet, magnesium sulfate, and other substances. Acids precipitate the casein by breaking up the combination with calcium phosphate. The action of rennet is more complex. Hammersten has shown that it is dependent upon the presence of calcium salts; thus, if the curd precipitated by dilute acid be dissolved in dilute alkali and the solution neutralized, it is unaffected by rennet, but regains its coagulability by the addition of a solution of a calcium salt or, what amounts to the same thing, a little of the whey from which the casein was precipitated. It appears that rennet splits the casein into two proteids, one of which is precipitated in the curd.

Halliburton uses the term "caseinogen" to designate the form in which the casein exists when in solution or precipitated by acids, and reserves the term casein for the curd produced by rennet.

Films of proteid matter occur abundantly in milk, for which reason it is distinctly opaque, even when all but a trace of the fat has been removed by centrifugal action.

The *albumin* of milk appears to be a distinct form, and is called lactalbumin. It is not precipitated by dilute acids, but is coagulated by heating to 70°–75° C. The proportion in cow's milk is usually from 0.35 to 0.50 per cent., but colostrum may contain much larger proportions.

Sébelien gives the composition of lactalbumin as follows:—

Carbon,	52.19
Hydrogen,	7.18
Nitrogen,	15.77
Oxygen,	23.13
Sulfur,	1.73

Globulin is present only in minute amounts in normal milk, but colostrum may contain as much as eight per cent. It is coagulated on heating.

Lactose.—This is the sugar of milk and is peculiar to it. Richmond discovered in the milk of the gamoose a sugar not identical with lactose; probably extended investigation of the milk of various animals will show the existence of other forms of milk-sugar. Crystallized lactose has the composition $C_{12}H_{22}O_{11} + H_2O$. By heating to 125° C. the water is expelled. It has a specific rotatory power of +52.5, which is independent of concentration. Freshly prepared solutions exhibit higher rotatory power, but after twenty-four hours standing, at ordinary temperatures, or immediately on boiling, the rotatory power is reduced to the figure given above. Alkaline solutions of copper salts are readily reduced by lactose at the boiling temperature. Richmond has shown that continued heating of a solution of milk-sugar causes caramelization and diminution of the rotatory power, but the reducing power to alkaline copper solution is not sensibly affected even after heating for two hours. In contact with yeast, lactose undergoes alcoholic fermentation, but with difficulty. It undergoes the lactic fermentation, however, very readily under the influence of certain microbes. When milk is evaporated rapidly to dryness, as in the determination of total solid residue, the milk-sugar is left in the anhydrous state.

Citric acid is a normal constituent of the milk of various animals. In human milk, the quantity is about 0.5 gram to the liter, in cow's milk from 1 to 1.5 grams. It is not dependent on the citric acid present in the food.

Minute amounts of nitrogenous bases and a starch-liquefying enzym also occur.

The ash of milk has the following composition:—

Ca,	.113	per cent.	(of milk)
Mg,	.0126	"	"
Fe,	.0002	"	"
K,	.146	"	"
Na,	.082	"	"
PO_4,	.263	"	"
SO_4,	traces	"	"
Cl,	.169	"	"
CO_3,	.020	"	"

Colostrum.—This term is applied to the milk secreted in the early stages of lactation. It usually differs markedly from ordinary milk. It contains characteristic structures known as colostrum corpuscles. These are ameboid bodies several times as large as the fat-globules. They are present for a variable period—three to fourteen days—but may persist much longer. Colostrum usually contains much less fat than fully developed milk, but a larger proportion of proteids, the increase being principally in the albumin, though Emmerling has found over 8 per cent. of globulin. The large proportion of albuminous matter causes colostrum to coagulate on boiling. Lactose is in small amount and is said to be largely replaced by another carbohydrate.

The following analysis is the average of the colostrum of 22 cows. (Eugling.)

Fat,	3.37
Casein,	4.83
Albumin,	15.85
Sugar,	2.48
Ash,	1.78
Total solids,	28.31

Colostrum is usually acid to litmus.

THE FOLLOWING TABLE IS A COMPILATION OF PUBLISHED ANALYSES OF VARIOUS MILKS.

	Human.	Cow.	Dog.	Elephant.	Mare.	Gamoose.	Goat.	Ass.	Ewe.	Sow.	Porpoise.
Specific Gravity, . . .					1034.9	1035.4	1032.9		1.0378		About 1.000
Fat, ...	3.8	3.9	9.37	20.58	1.09	5.56	4.3	1.6	9.50	4.8	45.80
Sugar, ...	6.00	4.7	3.09	7.18	6.65	5.41	4.0	6.1	5.00	3.4	1.33
Casein,	1.2	2.6	6.10	3.45	1.89	3.86	4.6	2.2	6.26	1.3	11.19
Albumin, .	0.5	0.6	0.50								
Ash, . .	0.2	0.7	0.73	0.65	0.31	1.03	0.6	0.5	1.01	0.9	0.57
Total Solids, .	11.7	12.5	19.79	31.86	9.94	15.86	13.5	10.4	21.77	10.4	58.89
Analyst, . . .	Lehmann and Hempel.		Koenig.	Koenig.	Vieth.	Pappel and Richmond.			Besana.		F. Purdie.

Normal milk is an opaque, white or yellowish-white fluid, with an odor recalling that of the animal, and a faint sweet taste. The opacity is due partly to the fat-globules, but when these are entirely removed the liquid does not become transparent. The reaction of freshly drawn milk is amphoteric to litmus, that is, it turns the red paper blue and blue paper red. The specific gravity varies between 1028 and 1035. It usually undergoes a gradual augmentation (sometimes termed Recknagel's phenomenon) for a considerable time after the sample has been drawn. The increase may amount to two units. The specific gravity becomes stationary in about five hours if the milk be maintained at a temperature below 15° C., but at a higher temperature it may require twenty-four hours to acquire constancy. The change is not dependent on the escape of gases, and is believed to be due to some molecular modification of the casein, possibly under the influence of an enzym.

Unless collected with special care and under conditions of extreme cleanliness, milk always contains bacteria and animal matter of an offensive character, such as epithelium, blood and pus cells, particles of feces and soil. Many minute organisms, especially bacteria, propagate with great rapidity in milk and produce changes in its composition. Some specific organisms, such as the *Spirillum choleræ*, multiply to only a limited extent in ordinary milk, being hampered by the bacteria normally present, but when introduced into sterilized milk increase with great rapidity.

At ordinary temperature milk soon undergoes decomposition under the influence of the microbes present, by which the milk-sugar is converted principally into lactic acid, and the proteids partly decomposed and partly coag-

ulated. The liquid becomes sour and the fat is enclosed in the coagulated casein.

In the initial stages of decomposition the proteids frequently undergo transformations into substances which are the cause of the violent poisonous effects occasionally produced by ice cream and other articles of food into the preparation of which milk enters.

Boiling produces coagulation of the albumin, some caramelization of the sugar, and develops a greater facility of coalescence on the part of the fat globules. Enzyms and most microbes are destroyed. The skin which forms on the surface of boiling milk is composed largely of casein. It is due probably to the more rapid evaporation at the surface of the liquid.

Partial freezing produces a concentration of the milk solids in the part remaining liquid, while the solid portion is deficient in them. The normal condition can, therefore, be restored only by thawing the entire mass and mixing thoroughly.

Richmond has published the following analyses as showing how great a difference may exist between the ice and liquid portion. The ice amounted to about 10 per cent.:—

	Ice.	*Liquid.*
Water,	96.23	86.62
Fat,	1.23	4.73
Sugar,	1.42	4.95
Proteids,	.91	3.90
Ash,	.21	.80
Specific gravity,	1009	1034.5

When milk is allowed to stand, some of the fat rises gradually and forms a rich layer, constituting cream. The proportion of cream depends on several conditions.

The amount formed in a given time cannot be taken as a measure of the richness of the milk. Water added to milk causes a more rapid separation of the cream. When milk is subjected to centrifugal action, a larger proportion of cream is quickly obtained, nearly all of the fat being removed. The following figures, given by D'Hout as averages, show the effect of the centrifugal action :—

	Whole milk.	*Separated milk.*	*Cream.*
Specific gravity, .	1032	1034	1015
Total solids, . .	14.10	9.6	26.98
Sugar, ...	4.70	5.05	3.32
Casein, . . .	3.50	3.62	2.02
Ash, ...	0.79	0.78	0 58
Fat, ...	5.05	0.20	21.95

Buttermilk is the residue after removal of the butter by churning. Vieth gives the following figures :—

Total solids.	*Fat.*	*Solids not fat.*	*Ash.*
9.03	0.63	8.40	0.70
8.02	0.65	7.37	1.29
10.70	0.54	10.16	0.82

The whey left after the precipitation of the curd by rennet or acid still contains a notable amount of proteids. The following analyses are by C. B. Cochran (*J. A. C. S.*, 1893, p. 347) :—

Milk		Whey.	
Total solids.	*Solids not fat.*	*Total solids.*	*Proteids removed.*
9.27	9.13	6.62	2.51
9.27	9.13	6.1	3.03
14.05	8.35	6.62	2.33
7.71	7.61	5.98	1.63
8.91	8.71	6.50	2.21

The whey of any given milk has the same composition, whether taken from the original milk, skimmed milk, or cream.

ANALYTIC PROCESSES.

The specific gravity determination is to be made only after the spontaneous rise has ceased. This will require about five hours, after the milk is drawn, if it has been kept below 15° C., but at a higher temperature it will be necessary to allow at least twelve hours. For all other determinations the milk must be analyzed as soon as possible, as it decomposes very rapidly at ordinary temperatures. The following figures, published by Bevan, show that a considerable loss in total solids may occur in twenty-four hours:—

	Total solids.	*Loss.*
Evaporated immediately, . . .	11.73	
" after 24 hours, . . .	10.79	0.94
" " 48 " . . .	10.38	1.35
" " 120 " . . .	9.42	2.31

The decomposition is very irregular and it is not possible to determine, by estimation of the lactic acid or other products, the original composition of the milk. The pipet used for taking a portion for analysis should have a wide opening, that no cream may be retained when the pipet is discharged.

SPECIFIC GRAVITY.

Air bubbles are held rather tenaciously by milk, and care must be taken in mixing, preparatory to taking the specific gravity, to avoid as far as possible the enclosure of

air, and to allow sufficient time for the escape of any bubbles that may be present. Determinations of specific gravity of milk are understood to be taken at the temperature of 60° F., and samples at temperatures materially different from this should be brought to it. If at a few degrees above or below 60°, it will suffice to take the gravity at once and obtain the correct figures by reference to Table A. The specific gravity of normal milk varies between 1028 and 1035. Since the specific gravity can be raised by the abstraction of fat (skimming) and restored by the addition of water, the figure taken alone cannot be relied upon as an index of the character of the sample, but in conjunction with the figure for fat or for total solids it is of great value, as a check on the results furnished by other determinations.

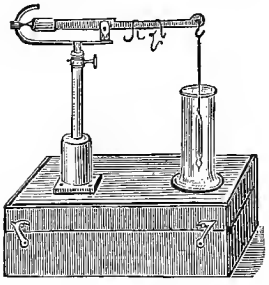
The simplest method of determining specific gravity is by the *lactodensimeter*, a delicate and accurately graduated hydrometer. The instrument must be immersed carefully so as not to wet the stem above the point at which it will rest. The instrument should be tested by immersion in distilled water at 15° C.

The indications furnished by the lactodensimeter are sufficiently accurate for most purposes, but its employment necessitates a considerable amount of the sample.

More accurate determination can be made by the *Westphal balance*. This is a delicate steelyard with a counterpoised plummet, displacing 5 c. c. The plummet being immersed in the milk, the equilibrium is restored by weights, the value of which can be directly expressed in figures for the specific gravity.

The principle of the Westphal balance may be applied by means of an analytic balance and a plummet. The

latter may conveniently consist of a short thermometer, or a thick glass rod, having a bulk of from 5 to 10 c. c. It is suspended from the hook of the balance by a fine platinum wire and its weight ascertained. It is then immersed in distilled water at 60° F. and the loss in weight noted. The figure so obtained is the weight of a bulk of water equal to that of the plummet. This having been determined, the specific gravity of a milk may be found by immersing the plummet in it and noting the loss in weight, which, divided by the loss in pure water, gives the specific gravity.



The *pyknometer* or *specific-gravity bottle* furnishes a means of accurate determination and is especially applicable when only a small amount of liquid is available. It is a small flask furnished with a perforated glass stopper. It is provided with a counterpoise equal to the empty flask and the weight of water that it carries is indicated. These data may, of course, be easily verified, and any error noted. The milk should be first brought to the proper temperature, the bottle completely filled, the stopper inserted, and the excess which flows out through the perforation and around the sides of the stopper removed by bibulous paper. The weight of milk divided by that of the water which the flask will hold at the same temperature gives the specific gravity.

TOTAL SOLIDS.

This determination is made by evaporation in a shallow, flat-bottomed dish, preferably of platinum, from 7 to 8 cm. in diameter. The milk must be spread evenly in a thin layer. If the ash is also to be determined, about 5 grams should be accurately weighed in the dish, evaporated rapidly to apparent dryness over the water-bath and the heating continued in the water-oven until the weight becomes practically constant, which will require about three hours. If the evaporation be slow, some decomposition occurs and the residue is brown, but if the larger portion of the water be evaporated quickly, a white residue is obtained. When the ash is not to be determined, it is more convenient to follow the method suggested by Richmond, using 1 to 2 grams, accurately weighed. The drying can be completed in about one and a half hours.

When a higher degree of accuracy is desired, Babcock's method (A. O. A. C.) may be employed as follows:—

Provide a hollow cylinder of perforated sheet metal 60 mm. long and 20 mm. in diameter, closed 5 mm. from one end by a disk of the same material. The perforations should be about 0.7 mm. in diameter and the same distance apart. Fill the cylinder loosely with from 1.5 to 2.5 grams of freshly ignited woolly asbestos free from fine or brittle material. Cool in a desiccator and weigh. Introduce a weighed quantity of milk (about 4 gm.) and dry at 100° C. to constant weight.

The residue will serve for the determination of the fat.

When rigid accuracy is not essential, it will suffice to measure the portion of milk taken for this and other determinations. Vieth uses a pipet graduated to deliver 5

grams and finds that, working with whole and skimmed milk, under the ordinary variations of temperature, the error will not exceed 0.1 on the total solids and is less on the fat.

A good plan is to use a 5 c. c. pipet and to wash out that which adheres to the glass with a little water. The specific gravity of the milk being known, the amount taken can be calculated. The milk should be as near 60° F. as possible.

ASH.

The residue from the determination of total solids is heated cautiously over the Bunsen burner, until a white ash is left. The result obtained in this manner is apt to be slightly low from loss of sodium chlorid. This may be avoided by heating the residue sufficiently to char it, extracting the soluble matter with a few c.c. of water, and filtering (using paper extracted with hydrofluoric acid). The filter is added to the residue, the whole ashed, the filtrate then added, and the liquid evaporated carefully to dryness. The ash of normal milk is about 0.7 per cent. and faintly alkaline; if the milk be watered the ash will be less. The ash of milk does not exactly represent the salts present in the milk. On ignition, the phosphorus of the casein is oxidized to phosphoric acid, which decomposes the carbonates formed by the ignition of the organic salts of the milk, and liberates carbonic acid. A marked degree of alkalinity and effervescence with hydrochloric acid will suggest the addition of a carbonate.

The method of the A. O. A. C. is as follows: In a weighed dish put 20 c.c. of milk from a weighing bottle, add 6 c.c. of nitric acid, evaporate to dryness, and burn at a low red heat until ~~all form~~ carbon.

the ash is free from

FAT.

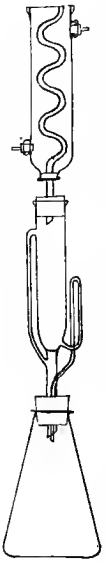
The method introduced by Wanklyn for the determination of fat by extracting it with ether from the total solid residue, has been found to give results 0.5 per cent. or more below the correct figure, and is therefore not described.

Adams' Method.—This consists essentially in spreading the milk over absorbent paper, drying, and extracting the fat in a Soxhlet apparatus; the milk is distributed in an extremely thin layer, and by a selective action of the paper the larger portion of the fat is left on the surface. It is essential that the paper contain no materials soluble in the liquid used for extraction. A paper, manufactured especially for this purpose by Schleicher & Schuell, is obtainable in strips of suitable size. Each of these yields to ether from .001 to .002 gram of extract, which may usually be disregarded.

The procedure is as follows: Five c.c. of the milk are discharged into a beaker 5 cm high and 3.5 cm. in diameter. The charged beaker is weighed, and a strip of the paper, which has been rolled into a coil, thrust into it. In a few minutes the paper will absorb nearly the whole of the milk. The coil is then carefully withdrawn, and stood dry end downward on a sheet of glass.

With a little dexterity, all but the last fraction of a drop will be absorbed by the paper. The beaker is again weighed and the milk taken found by difference. It is of importance to take up the whole of the milk from the beaker, as the paper has a selective action, removing the watery constituents by preference over the fat. The charged paper is placed in the water-oven on a glass plate,

milk-end upward, and dried. Usually about one hour is sufficient. It is inserted in a Soxhlet continuous-extraction apparatus, the tared flask of which should have a capacity of about 150 c. c. and contain about 75 c. c. of anhydrous, alcohol-free ether, or petroleum spirit boiling at about 45° C. Heat is applied to the flask by means of a water bath, or by resting it on a piece of asbestos paper, which is heated by a small flame. After the coil has received at least ten or twelve washings, the flask is detached, the ether removed by distillation, and the fat dried by heating in an air oven at about 105° C., and occasionally blowing air through the flask. After cooling, the flask is wiped with a piece of silk, allowed to stand ten minutes, and weighed.



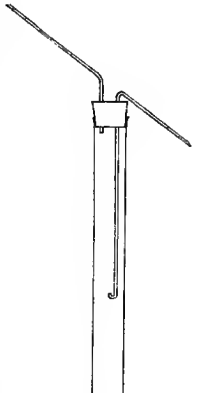
Richmond states that to perform a rigidly accurate determination attention to the following points is necessary: The ether must be anhydrous (drying over calcium chlorid and distilling is sufficient). Schleicher & Schuell's fat-free papers should be used and one should be extracted without any milk on it, as a tare for the others. Four or five hours' extraction is necessary, and the coils should be well dried before extraction is begun.

Thimble-shaped cases made of fat-free paper are now obtainable and are convenient for holding the absorbent material on which the milk is spread. The fine texture prevents undissolved matter escaping. A case may be used repeatedly. Sour milk must be thinned with ammonium hydroxid before taking the portion for analysis.

When the Babcock method has been used for determining the total solids, the cylinder and contents may be placed in the Soxhlet tube and extracted with ether in the usual way. In addition to weighing the fat, after evaporation of the ether, it may be determined by difference, by drying the extracted cylinder at 100° C. and weighing. A still higher accuracy is secured by performing all the drying operations in an atmosphere of hydrogen.

Werner-Schmid Method.—This is a very satisfactory and rapid method for the determination of fat and is especially suitable for sour milk.

Ten c. c. of the milk are measured into a long test-tube of 50 c. c. capacity, graduated to tenths c. c., and 10 c. c. of strong hydrochloric acid added, or the milk may be weighed in a small beaker and washed into the tube with the acid. After mixing, the liquid is boiled one and a half minutes, or the tube may be corked and heated in the water bath from five to ten minutes, until the liquid turns dark brown. It must not be allowed to turn black. The tube and contents are cooled in water, 30 c. c. of *well-washed* ether added, shaken, and allowed to stand until the line of acid and ether is distinct. The cork is taken out, and a double-tube arrangement, like that of the ordinary wash-bottle, inserted. The stopper of this should be of cork and not of rubber, since it is difficult to slide the glass tube in rubber, and



there is a possibility, also, of the ether acting on the rubber and dissolving it. The lower end of the exit-tube is adjusted so as to rest immediately above the junction of the two liquids. The ethereal solution of the fat is then blown out and received in a weighed flask. Two more portions of ether, 10 c. c. each, are shaken with the acid liquid, blown out, and added to the first. The ether is then distilled off, and the fat dried and weighed as above.

Leffmann-Beam Method.—Among the processes for the rapid determination of fat those employing centrifugal machines have been most satisfactory. The following method can be applied very cheaply, both as regards the original cost of the apparatus and of the chemicals required for the test, the latter being usually no inconsiderable item when many tests are made. The manipulation is very simple.

Machines arranged for either two, four, six, eight, or twelve bottles are manufactured. The process is covered by patent in the United States, which has been assigned to J. E. Lonergan, of Philadelphia.

The test-bottles have a capacity of about 30 c. c., and are provided with a graduated neck, each division of which represents one-tenth per cent. by weight of butter-fat.

Fifteen c. c. of the milk are measured into the bottle, 3 c. c. of a mixture of equal parts of amyl alcohol and strong hydrochloric acid added, mixed, the bottle filled nearly to the neck with concentrated sulfuric acid and the liquids mixed by holding the bottle by the neck and giving it a gyratory motion. The neck is now filled to about the zero point with a mixture of sulfuric acid and water prepared at the time. It is then placed in the centrifugal machine, which is so arranged that when at rest the bot-

tles are in a vertical position. If only one test is to be made, the equilibrium of the machine is maintained by means of a test-bottle, or bottles, filled with a mixture of equal parts of sulfuric acid and water. After rotation for from one to two minutes, the fat will collect in the neck of the bottle and the percentage may be read off. It is convenient to use a pair of dividers in making the reading. The legs of these are placed at the upper and lower limits respectively of the fat, allowance being made for the meniscus; one leg is then placed at the zero point and the reading made with the other. Several years' experience by analysts in various parts of the world has shown that with properly graduated bottles the results are reliable. As a rule, they do not differ more than one-tenth of 1 per cent. from those obtained by the Adams process, and are generally even closer. Richmond, as the result of an exhaustive series of investigations, draws the following conclusions:—

1. As variations in the sulfuric acid and fusel-oil may slightly influence the result, it is well to obtain large quantities of these at a time, and by a few preliminary experiments fix the factor necessary to convert scale readings into percentages of fat.

2. It is advisable to use the same strength of acid (94 to 96 per cent. H_2SO_4 is convenient). This may be estimated either by converting a known quantity into ammonium sulfate, drying at 100° C., and weighing; titrating the acidity with decinormal barium hydroxid, using methyl-orange as indicator, and then igniting, and weighing the non-volatile matter (correcting for the barium sulfate formed); or deducing the strength from the density (hydrometers are rarely of sufficient accuracy, and a pyc-

nometer should be used), and correcting by subtracting twice the weight of the non-volatile matter.

3. If it be necessary to perform the experiment when the atmospheric temperature is high, the milk and acid should be cooled beforehand, or if this is not practicable, the acid should be added in small portions (2 c. c.) at a time, and the bottle shaken between each addition, or a weaker acid may be used.

4. About half to three-quarters of a minute's revolution, at the rate of from 1200 to 1500 revolutions a minute, has been found the most convenient in practice. The machine has not been found dangerous at this speed; but, if wished, a longer period of revolution at a slower speed is equally efficacious.

For accurate work the factor for correcting the reading on each of the bottles should be determined by comparison with the figures obtained by the Adams or other standard process.

Cream is to be diluted to exactly ten times its volume, the specific gravity taken, and the liquid treated as a milk. Since in the graduation of the test-bottles a specific gravity of 1030 is assumed, the reading must be increased in proportion.

A more accurate result may be obtained by weighing in the test-bottle about 2 c. c. of the cream and diluting to about 15 c. c. The reading obtained is to be multiplied by 15.45 and divided by the weight in grams of cream taken.

The mixture of amyl alcohol and hydrochloric acid seems to become less satisfactory when long kept. It is best, therefore, not to make up large amounts at once. The mixture should be clear and not very dark in color.

Samples of amyl alcohol that produce a black solution with hydrochloric acid are unsuitable. (See Appendix.)

Calculation Method.—Several investigators have proposed formulæ by which when any two of the data, specific gravity, fat, and total solids are known, the third can be calculated. Since fairly accurate determinations of the fat and specific gravity can be made by rapid and simple methods, the formulæ become very serviceable and should always be used to check the results. That of Hehner and Richmond has been almost exclusively used.

It is as follows :—

$$F = .859\ T - .2186\ G$$

in which F represent the fat, T the total solids, and G the specific gravity. This formula will suffice for ordinary milks, but for poor skim milks it has been found necessary to modify it as follows:—

$$F = .859\ T - .2186\ G - .05\ (\frac{G}{T} - 2.5)$$

This correction is to be applied only when G divided by T exceeds 2.5.

In these formulæ, G represents the last two units of the specific gravity and any decimal. Thus, if the observed specific gravity be 1029.5, G will be 29.5.

A ready means of applying the formula is by the use of Richmond's slide-rule. This has three scales, two of which, for fat and total solids respectively, are marked on the body of the rule, while that for the specific gravity is placed on the sliding portion.

The divisions are as follows :—

Total solids, ... 1 inch divided into tenths
Fat, ... 1.164 inches " " "
Specific gravity, . . . 0.254 " " " halves

The rule is employed by adjusting the arrow point to the graduation corresponding to the fat found, when the figure for the total solids will coincide with that for the observed specific gravity. This instrument does not take into consideration the necessary correction for poor skim milks, but the error from this cause will in no case exceed .08 per cent. of fat and may usually be disregarded.

It has been found that the fat calculated by the above formula is slightly higher than the actual amount. Recently, Richmond has proposed a new formula—

$$T = .2625 \left(\frac{G}{\frac{1000 + G}{1000}}\right) + 1.2\ F$$

and finds the results obtained by it to agree much better with accurate analysis.

The formula may be simplified as follows,

$$T = \frac{G}{4} + \frac{6}{5} F + 1.4$$

and the result will not differ in extreme cases more than .02 from those calculated from the more complex formula. Results calculated from this formula will be found in Table B.

A milk-scale to express the same relation may be constructed on which 1 per cent. total solids = 1 inch, 1 per cent. of fat = 1.2 inches, and 1 degree of gravity = $\frac{1}{4}$ inch; if the zero on the fat scale be placed on a line with 5 per cent. on the total-solids scales, the arrow will be in its correct position: 0.14 ($\frac{1}{7}$) inch below 20 degrees on the specific gravity scale. By placing an arrow 0.14 inch below the present arrow, existing milk scales will give a near approximation.

TOTAL PROTEIDS.

This determination is best made by calculation from the figure for total nitrogen obtained by Gunning's modification of Kjeldahl's process. The reagents and apparatus required are as follows:—

Standard sulfuric acid.—This should be about decinormal, the exact value being determined by precipitating a measured volume with barium chlorid, collecting and weighing the barium sulfate under the usual precautions.

Standard barium hydroxid.—This should be about decinormal. The volumetric relation between this solution and the standard sulfuric acid must be accurately determined, employing phenolphthalein as an indicator.

Acid potassium sulfate mixture.—One part of pure potassium sulfate is heated with two parts of sulfuric acid (strictly C. P.) until the potassium sulfate is dissolved. The mixture is semi-solid when cold but may be readily liquefied by warming.

Sodium hydroxid solution.—A saturated solution in water. A good quality of pulverized sodium hydroxid is now sold under various trade names, *e. g.*, "Lewis' Lye," "Banner Lye," and these will be found satisfactory.

Digestion and distillation flask.—This has a body capable of holding about 550 c. c., and a cylindrical neck about 18 cm. in length and 25 cm. in diameter. It is supported on wire-gauze or asbestos, and the mouth may be covered by a watch-glass or funnel during the digestion. For distilling, a well-fitting rubber stopper with delivery tube should be attached. The tube should be of the same diameter as the condensing tube, and should have one or two bulbs, about 4 cm. in diameter, to prevent any solution

being carried over by spurting. It should project slightly below the stopper and be cut off obliquely.

Condenser.—The condensing tube must be of block tin of an external diameter of about one centimeter. At least 30 centimeters of its length should be in contact with the cooling water. The junction of the glass and tin tube is made by a short, close-fitting rubber tube, and the tubes are so bent as to slope toward the distilling flask. The lower end of the tin tube is connected by a short rubber tube with a glass bulb-tube which dips below the surface of a measured volume (20 c. c.) of the standard sulfuric acid in an Erlenmeyer flask of about 300 c. c. capacity.

Five c. c. of the milk are weighed or measured into the flask and evaporated to dryness over the water-bath ; 30 c.c. of the acid potassium sulfate mixture are added and heated over the Bunsen burner. At first, frothing occurs and white fumes escape, consisting chiefly of water-vapor. To prevent loss of acid, the neck of the flask is now fitted with a funnel which is covered with a watch glass. This will cause the acid to condense and run back into the flask. The operation is finished when the liquid is colorless, which generally requires about an hour. When cold, the contents of the flask are diluted with about 200 c. c. of water, several pieces of ignited pumice dropped in, and sufficient of the sodium hydroxid solution (about 50 c. c.) added to make the mixture strongly alkaline. It should be poured down the side of the flask so that it does not mix at once with the acid. The flask is now connected with the condenser and the contents mixed by shaking. The liquid is now distilled until the whole of the ammonium hydroxid is collected, which will usually be the case when 150 c. c. have passed over. The receiver and short tube dipping in

it are then detached and the distillate titrated to determine the amount of acid unneutralized. From this the amount of ammonium hydroxid can be calculated, and the nitrogen in this multiplied by 6.38 will give the figure for the total proteids.

The A. O. A. C. recommends an indicator prepared as follows: Three grams of cochineal are digested for several days at room temperature in a mixture of 50 c. c. of strong alcohol and 200 c. c. of water. The filtered solution is used.

The Ritthausen Method for Total Proteids.—This method depends on precipitation by copper sulfate and sodium hydroxid. It is applicable only to fully developed milks; the proteids of colostrum and whey are only partially precipitated. The reagents are:—

Copper sulfate solution.—34.639 grams of pure crystallized copper sulfate are dissolved and made up to 500 c. c.

Sodium hydroxid solution.—About 12 grams are dissolved in 500 c. c. of water.

Ten grams of the milk are placed in a beaker, diluted with 100 c. c. of distilled water, 5 c. c. of copper sulfate solution added, and thoroughly mixed. The sodium hydroxid solution is then added drop by drop, with constant stirring, until the precipitate settles quickly and the liquid is neutral, or at most very feebly acid. An excess of alkali will prevent the precipitation of some of the proteids.

The reaction should be tested on a drop of the clear liquid, withdrawing it by means of a rod, taking care not to include any solid particles. When the operation is correctly performed, the precipitate, which includes the fat, settles quickly, and carries down all of the copper.

It is washed by decantation with about 100 c. c. of water, and collected on a filter (previously dried at 130° C. and weighed in a weighing bottle). The portions adhering to the sides of the beaker are dislodged with the aid of a rubber-tipped rod. The contents of the filter are washed with water until 250 c. c. are collected, which are mixed and reserved for the determination of the sugar as described below. The water in the precipitate is removed by washing once with strong alcohol, and the fat by six or eight washings with ether. The Soxhlet apparatus may be used for this purpose. The washings being received in a weighed flask, the determination of the fat may be made by evaporating the ether with the usual precautions.

The residue on the filter, which consists of the proteids in association with copper hydroxid, is washed with absolute alcohol, which renders it more granular, and then dried at 130° C. in the air bath. It is weighed in a weighing bottle, transferred to a porcelain crucible, incinerated, and the residue again weighed. The weight of the filter and contents, less that of the filter and residue after ignition, gives the weight of the proteids. The results by this method are slightly high, since copper hydroxid does not become completely converted into copper oxid at 130° C.

Richmond and Boseley (*Analyst*, 1893) modify this process by diluting the milk to 200 c. c., adding a little phenolphthalein, and neutralizing any acidity by the cautious addition of dilute sodium hydroxid solution, then adding from 2.0 to 2.5 c. c. of the copper sulfate solution. The precipitate is allowed to settle, washed, and estimated as above.

CASEIN AND ALBUMIN.

Twenty c. c. of the milk are mixed with 40 c. c. of a saturated solution of magnesium sulfate and powdered magnesium sulfate stirred in until no more will dissolve. The precipitate of casein and fat, including the trace of globulin, is allowed to settle, filtered, and washed several times with a saturated solution of magnesium sulfate. The filtrate and washings are saved for the determination of albumin. The filter and contents are transferred to a flask and the nitrogen determined by the method described above. The nitrogen so found multiplied by 6.38 gives the casein.

The filtrate and washings from the determination of casein are mixed, the albumin precipitated by a solution of tannin, filtered, and the nitrogen in the precipitate determined as above. The same factor is used.

On account of the difficulty of washing the precipitated casein, we prefer to proceed as follows: 20 c. c. of the milk are mixed with saturated magnesium sulfate solution and the powdered salt added to saturation. The mixture is washed into a graduated measure with a small amount of the saturated solution, mixed, the volume noted, and allowed to stand until the separation takes place. As much as possible of the clear portion is drawn off with a pipet and passed through a dry filter. An aliquot portion of the filtrate is taken, the albumin precipitated by a solution of tannin, and the nitrogen in the precipitate determined as above.

The casein is found by subtracting the figure for albumin from that for total proteids.

For the separation of the proteids in milk, the methods of Hoppe-Seyler and Ritthausen have been much used.

The casein is precipitated by the addition of acetic acid to the diluted milk, the action being rendered complete, in the one case, by raising the temperature to 40° C., and in the other by passing a current of carbon dioxid through the mixture at ordinary temperatures. L. L. Van Slyke (*J. A. C. S.*, Vol. xv, p. 635), as the result of a series of investigations, finds these methods to give practically identical results and recommends the following procedure for the separation and estimation of the casein and albumin :—

Casein.—Weigh out about ten grams of milk, dilute in a beaker with about 90 c. c. of water at 40°–42° C., and add at once 1.5 c. c. of a solution containing 10 per cent. of acetic acid by weight. Stir with a glass rod, let stand three to five minutes, decant on to a filter, wash two or three times with cold water by decantation, transfer the precipitate completely to the filter, and wash it once or twice. The washed precipitate and filter paper are then treated by the Kjeldahl–Gunning method for the determination of nitrogen, and the estimation made in the usual manner. To calculate the nitrogen into an equivalent amount of casein, multiply by the factor 6.38.

Albumin.—The filtrate from the separated casein is heated in boiling water for ten or fifteen minutes. The coagulated albumin is collected in a filter, washed, and the filter and precipitate treated by the Kjeldahl–Gunning method. The amount of nitrogen multiplied by 6.38 gives the amount of albumin.

Remaining nitrogen compounds.—The remaining compound or compounds of nitrogen are determined by difference, subtracting from the amount of total nitrogen compounds the sum of the casein and albumin.

The albumin of condensed milk is partly coagulated by the heat employed in its manufacture. When magnesium sulfate is added, therefore, to precipitate the casein, the coagulated albumin will be carried down at the same time

and only the soluble albumin will be found in the filtrate. Faber (*Analyst*, 1889) has applied this fact to the detection of the previous heating of a sample of milk. Usually, only about one-third of the albumin is found uncoagulated in condensed milk.

SUGAR.

The following method, due to Soxhlet, employs a Fehling's solution, made as required, by mixing equal parts of the following solutions:—

Copper sulfate solution.—34.639 grams of pure crystallized copper sulfate are dissolved in distilled water and made up to 500 c. c.

Alkaline tartrate solution.—One hundred and seventy-three grams of pure sodium potassium tartrate, and 51 grams of sodium hydroxid of good quality, are dissolved and made up to 500 c. c.

One hundred c. c. of the mixed filtrate obtained as described on p. 33 are brought to boiling, 50 c. c. of boiling Fehling's solution added, and the boiling continued for six minutes. The precipitate is allowed to settle for a short time, and the supernatant liquid poured through a filter. About 50 c. c. of boiling water are added to the residue, and the heating continued for a minute or two. The precipitate is then conveyed to the filter, washed with boiling water, with alcohol, and finally with a small quantity of ether. The filter and contents are dried in the water oven, the precipitate removed to a tared porcelain crucible, the filter held over the crucible and burnt to ash, which is added to the precipitate, and the cuprous oxid converted into cupric oxid by strong ignition, for five or ten minutes, over the Bunsen burner.

The amount of copper reduced under the conditions detailed above is not directly proportional to the milk-sugar present. Table C shows the amounts of milk-sugar ($C_{12}H_{22}O_{11} + H_2O$) equivalent to given weights of cupric oxid. The volumes of Fehling's solution and sugar solution must conform strictly to the figures given above.

Many of the English analysts prefer to make volumetric determinations by means of Pavy's modification of Fehling's solution, in which strong ammonium hydroxid is used to maintain the cuprous oxid in solution and thus permit nicer determination of the end-reaction. Allen has investigated with great care the value of this method for determining the form of glucose that occurs in urine and finds that considerable variation may be made in the formula of Pavy's solution without the oxidizing ratio being appreciably affected, but it will be best to adhere closely to a particular formula in milk determinations. A description of the process is given in connection with analysis of condensed milks.

The determination of sugar may also be made by means of the polarimeter after removal of the fat and proteids. This may be effected by means of a nitric acid solution of mercuric nitrate as suggested by Wiley.

The mercuric nitrate solution is prepared by dissolving mercury in twice its weight of nitric acid of 1.42 sp. gr. and adding to the solution five volumes of water.

Sixty c. c. of the milk are placed in a 100 c. c. flask and 10 c. c. of the mercuric solution added. The flask is filled to the mark with water, well shaken, and the liquid filtered through a dry filter. The filtrate, which will be perfectly clear, may be examined at once in the polarimeter. Several readings should be made and the average taken.

It is to be noted that the actual volume of the sugar-containing solution is 100 c. c., less the space occupied by the precipitated proteids and fat. The volume of fat is found by multiplying the weight in grams by 1.075 and the proteids by multiplying the weight by 0.8.

For example :—
Sp. gr. of milk 1030, fat 4 per cent., proteids 4 per cent.
Milk taken = 60 × 1.03 = 61.80 gms.
The weight of fat = 4 per cent. of 61.80 = 2.47 gms.
The weight of proteids = 4 per cent. of 61.80 = 2.47 gms.
The volume of fat = 2.47 × 1.075 = 2.65 c. c.
The volume of proteids = 2.47 × .8 = 1.97 c. c.
The bulk of the sugar containing liquid is therefore
100 — (2.65 + 1.97) = 95.38 c. c.

In order to avoid the calculation involved in taking 60 c. c. of the milk as given above, an amount may be employed which is a simple multiple of the standard quantity to be used in the polarimeter at hand. Thus, for instruments adjusted so that 16.19 grams of sucrose (20.56 grams of milk-sugar) in 100 c. c. of the solution produce a rotation of 100 degrees on the per cent. scale, 61.68 grams (20.56 × 3) may be weighed out directly for the purpose and made up to 100 c. c. plus the volume occupied by the fat and proteids, the latter being calculated as above. The sugar-containing liquid will then be exactly 100 c. c., and the reading on the polarimeter divided by three will give the percentage of hydrated milk-sugar direct if a 200 mm. tube be employed. With a 400 mm. tube or 500 mm. tube the reading is to be divided by 6 or 7.5 respectively.

Polarimeters.—A discussion of the construction of the various forms of polarimeters and of the optical principles

involved, would be beyond the scope of this work. The most economic instruments are the so-called half-shadow instruments, using the sodium flame, and they are the most satisfactory. They are so arranged that the field is divided into semicircles which are equally illuminated when the instrument registers zero. On the introduction of the tube carrying the sugar solution, the illumination becomes unequal and the angular rotation of the analyzer, which is required to restore the original condition, measures the rotation which has been caused by the sugar. Most instruments are furnished with two scales, one showing the rotation in angular degrees and the other expressing per cent. directly. The latter reads to 100 when a certain fixed quantity of the material has been dissolved in water and diluted to 100 c. c.

The *specific rotatory power* of a substance is the amount of rotation of the plane of polarized light, in angular degrees, produced by a solution containing one gram of the substance in 1 c. c., examined in a column 1 decimeter long.

It is expressed by the following formula, in which

S is the specific rotatory power for light of wave length corresponding to the D line of the spectrum (sodium flame).

a is the angular rotation observed,

c is the concentration of the solution (weight in grams, in 100 c. c. of the liquid), and

l is the length of the tube in decimeters.

$$S = \frac{100\ a}{cl}$$

Calculation of the amount of sugar corresponding to the observed rotation may be made by substitution in the formula.

The specific rotatory power of milk-sugar is unaffected by the concentration within the limits encountered in ordinary milk-analysis. It is slightly affected by temperature, being decreased by about .075 angular degree for each successive rise of one degree Centigrade. The specific rotatory power at 20° C. is 52.5° when observed by the sodium flame.

The employment of an arbitrary factor for correcting for the volume of precipitate may be avoided by the so-called method of "double dilution," in which two solutions of different volume are compared. With the higher class of polarimeters, the determinations may be easily made within one-tenth per cent. The following is a summary of the method given by Wiley and Ewell (*J. A. C. S.*, 1895, p. 428) who recommend it strongly. The instrument used was the new triple-field shadow polarimeter made by Schmidt and Haensch, which permits readings to be made within 0.05 per cent. 32.91 grams of lactose dissolved in 100 c. c. gave a reading of 100. The amount of milk taken was double this quantity, that is, 65.82 grams, which were placed in a 100 c.c. flask, 10 c.c. of the acid mercuric nitrate added, the flask filled to the mark, the contents well mixed, filtered, and polarized. A similar quantity of the milk was placed in a 200 c. c. flask and treated in the same way. The true polarization is obtained by dividing the product of the readings in the two flasks by their difference. Thus in the paper above noted the following experiments are recorded:—

Reading in 200 c. c. flask.	*Reading in 100 c. c. flask.*	*Apparent per cent. lactose.*	*True per cent. lactose.*
10 15	20.84	5.21	4.95

The polarimeter used had a tube 4 decimeters long.

The figure for apparent percentage is obtained by dividing the reading of the small flask by 4. The true percentage is obtained by multiplying 10.15 by 20.84, dividing by their difference (10.69), and taking one-fourth this quotient.

Birotation.—When freshly dissolved in cold water, milk sugar shows a higher rotation than that given above. By standing, or immediately on boiling, the rotatory power falls to the point mentioned. In preparing solutions from the solid milk-sugar, care must be taken to bring them to the boiling point previous to making up a definite volume. This precaution is unnecessary when operating upon milk.

MILK ADULTERANTS.

Water.—The addition of water to milk is usually detected by the diminution in the amount of solids. It was formerly supposed that normal milk does not contain nitrates, and since these are almost universally present in surface and subsoil waters, it was suggested that the application of some of the delicate tests for nitrates would detect the addition of water, but Bevan has shown that pure milk may contain nitrates, and Richmond obtained a reaction for nitrates from the milk of a cow to which had been administered a very small quantity of niter.

The addition of water decreases the specific gravity, while abstraction of fat increases it. It is possible, therefore, by carrying out both methods of adulteration carefully, to maintain the same gravity as in the original sample, so that this datum alone will not suffice to detect adulteration. Taken in conjunction with either the figure for fat or for total solids, the specific gravity becomes of

direct value, and furnishes a means for determining, by calculation, the remaining datum.

Vieth has pointed out that in normal milks the following ratios obtain: Sugar : proteids : ash = 13 : 9 : 2, and a determination of these ratios may aid in the attempt to distinguish genuine but abnormal milks from watered milks. In the case of a watered milk the proportion would remain unchanged, but in abnormal milk it has been found to vary. Richmond finds that "the most constant figure in normal milks is the proportion of ash to solids-not-fat, which averages 8.3 per cent. and very rarely passes outside of the limits of 8.0 per cent. and 8.5 per cent. In cases of low solids-not-fat this proportion has been disturbed, and the ash has had a higher ratio to the solids-not-fat. In no case has the percentage of ash in the milk fallen below 0.7 per cent., even in milks notably below the limit. In an adulterated milk containing, say, 8 per cent. of solids-not-fat, the ash would usually be lower than this—about 0.66 per cent.; this difference is small, but as the ash is capable of being estimated with great accuracy, it is significant. Other observers have found the same thing."

For milk control in dairies, etc., it will suffice to take the specific gravity by the lactodensimeter (see page 19) and the fat by the Leffmann-Beam method. From the figures thus obtained the total solids can be ascertained by Table B or Richmond's slide-rule.

Various substances are added to milk to conceal adulteration or inferiority in quality. The most frequently employed are coloring matters. Sugar, salt, and starch have been added to milk, but are of infrequent occurrence. It has been stated that chalk has been added, but this is ob-

viously unlikely. The coloring matters most frequently employed are annatto and certain coal-tar colors. Caramel, saffron, and carotin are occasionally used.

Annatto is easily detected by rendering the sample slightly alkaline by the addition of sodium acid carbonate, immersing in it a slip of filter paper, and allowing it to remain over night. The presence of annatto will be indicated by a distinct reddish-yellow tinge to the paper.

Coal-tar colors are detected by adding to the milk ammonium hydroxid and allowing a piece of white wool to remain in it over night. The dye is taken up by the wool, which acquires a yellow tinge. When milk contains Martius' yellow, ammonium hydroxid intensifies the color and hydrochloric acid bleaches it.

Further information as to coloring matters will be found under the analysis of butter.

Starch may be detected by the blue color developed on the addition of solution of iodin to the milk, which has previously been heated to the boiling temperature and then cooled. Starch is very often added to ice-cream and similar articles.

Salt and **cane sugar** are occasionally added to milk that has been diluted with water. The former is easily detected by the taste, the increased proportion of ash and of chlorin. Cane sugar may be detected, if in considerable quantity, by the taste. The quantitative determination is made by the methods described in connection with condensed milk.

Antiseptic substances are largely used, especially in the warmer season, as a substitute for refrigeration. Many of these are sold under proprietary names which give no indi-

cation of their composition. Preparations of boric acid and borax were at one time the most frequent in use, but lately, formalin, a 40 per cent. solution of formaldehyde (methyl aldehyde), has come into favor. Sodium benzoate is now in common use as a preservative for cider, fruit-jellies, and similar articles, and may, therefore, be found in milk. Salicylic acid is not so much employed as in former years. Sodium carbonate is occasionally used to prevent coagulation due to slight souring.

R. T. Thomson ("Glasgow City Anal. Rep., 1895," quoted in *Analyst*, March, 1896) has studied the comparative value of milk preservatives. He finds that a mixture of boric acid and borax is more efficient than the acid alone. The quantity generally used is equivalent to about one-half gram of boric acid per liter. Formalin was shown to be by far the most efficient antiseptic. In the proportion of 0.125 gram to the liter, it kept milk sweet for eight days.

Formaldehyde.—The presence of this body may sometimes be detected by its odor developed on warming the milk. Hehner's method, the most characteristic for its detection, depends upon the fact that when milk containing it is mixed with sulfuric acid a blue color appears. Richmond and Bosely showed that the delicacy of the test is much increased by diluting the milk with an equal bulk of water and adding sulfuric acid of 90 to 94 per cent., so that it forms a layer underneath the milk. Under these conditions, milk, in the absence of formaldehyde, gives a slight greenish tinge at the junction of the two liquids, while a violet ring is formed when formaldehyde is present even in so small a quantity as one part in 200,000 of milk. The color is permanent for two or three days. In the absence of formaldehyde, a brownish color is developed after

some hours, not at the junction of the two liquids but lower down in the acid.

Another test is by the use of Schiff's reagent—a solution of magenta bleached by sulfurous acid. In presence of an aldehyde a pink color is developed. This test is usually applied to the distillate from the milk, but Richmond and Bosely consider it safe to use the whey produced by adding dilute sulfuric acid to the milk. They point out further that great care must be exercised in applying the test. Excess of sulfurous acid must be carefully avoided in preparing the reagent, since the color is not developed in its presence. On the other hand, a red color is easily developed in the reagent by warming, blowing air through it, or even placing it in an uncorked bottle. Hehner has shown that when a small amount of the reagent, say two drops, is added to the distillate from the milk, a red color, due to the oxidation of the sulfurous acid by the oxygen of the water, is developed after some time, whereas no color appears if a larger amount—say ten drops—has been added. He recommends to add about five drops of the reagent to the distillate (amounting to about 25 c.c.) from 100 c.c. of milk, place the mixture in a stoppered cylinder, observe the color next morning, and then add a few drops of sulfurous acid solution. After a short time, any color which may be due to oxidation will have vanished, while that due to the presence of an aldehyde remains. There is a difference in the tint produced by color oxidation, which resembles that of rosanilin, and that of the aldehyde compound, which is violet; and with the small amounts that are often to be detected, only a comparison of the relative colors would allow of anything like a safe conclusion being drawn. Hehner also recommends the following as a sensi-

tive and characteristic test: To the distillate from the milk, add one drop of a dilute aqueous solution of phenol, pour the mixture upon strong sulfuric acid contained in a test-tube. A bright crimson zone appears at the line of contact. This color is readily seen with one part of formaldehyde in 200,000 of water. If there is more than one part in 100,000, there is seen above the red ring a white, milky zone, while in stronger solutions a copious white or slightly pink, curdy precipitate is obtained. This reaction has an advantage over the one above referred to, in that it is obtained with formaldehyde solution of all strengths, while the blue color with sulfuric acid is not obtained with milk containing much formaldehyde. Acetaldehyde also gives a coloration and a precipitate with phenol and sulfuric acid, but it is orange-yellow, not crimson.

Many hydroxy-derivatives of benzene, such as salicylic acid, resorcinol, and pyrogallol, give the red color with formaldehyde. Hydroquinone does not give the red color but only an orange-yellow one.

The reaction only succeeds when carried out as described above; the phenol must first be mixed with the solution to be tested, and the mixture poured upon the sulfuric acid. Only a trace of phenol must be used. If the hydroxy-compound is first dissolved in the acid and the formaldehyde solution then added, no color is obtained.

The precipitate obtained by sulfuric acid, formaldehyde, and phenol is highly insoluble, and might be utilized for the determination of the strength of dilute formalin solutions.

Benzoates.—Two hundred and fifty to five hundred c. c. of the sample are rendered alkaline by a few drops of calcium or barium hydroxid, evaporated to one-fourth bulk,

mixed with sufficient calcium sulfate, powdered pumice, or other inert material as an absorbent, to make a pasty mass, and dried on the water bath. When condensed milk is examined, 100 to 150 grams should be mixed directly with sufficient absorbent material and a few drops of barium hydroxid. The dry mass is finely powdered, moistened with dilute sulfuric acid, and then exhausted three or four times with about twice its volume of cold (50 per cent.) alcohol, which dissolves benzoic acid freely, but only mere traces of the fat. The alcoholic liquid which, in addition to the benzoic acid, contains milk-sugar and mineral matter, is mixed thoroughly, neutralized with barium hydroxid, and evaporated to small volume. The residue is acidified with weak sulfuric acid and extracted with successive small portions of ether. On evaporation, the ether leaves, almost pure, the benzoic acid, which may be recognized by its odor and volatility.

Boric Acid.—When boric acid or borates are not present in quantities sufficient to appreciably increase the ash of the sample, the quantitative determination is difficult. The qualitative test is delicate. One hundred c. c. of the sample are rendered alkaline with calcium hydroxid, evaporated and ashed. Calcium hydroxid is preferred because the ashing takes place more rapidly. The ash is dissolved in the smallest possible quantity of strong hydrochloric acid, the solution filtered, and evaporated to dryness. An appreciable loss of boric acid will not occur. The residue is moistened with very dilute hydrochloric acid, mixed with tincture of turmeric, and dried on the water bath. The smallest trace of boric acid gives to the residue a vermilion or cherry-red tint.

Concentrated hydrochloric acid gives, with tincture of

turmeric, a cherry-red color, which, however, disappears on addition of water, and also becomes brown on drying, while the boric acid color appears only on drying, and is not destroyed unless much water be added, or at the boiling point. The red color adheres strongly to the vessel, and may be removed by alcohol. The flame test may be applied to the residue, but it is not delicate.

R. T. Thomson (*Analyst*, March, 1896) gives the following method for the estimation of boric acid. One hundred c. c. of milk are mixed with from one to two grams of sodium hydroxid, evaporated to dryness in a platinum dish, the residue thoroughly charred, heated with 20 c. c. of water, and hydrochloric acid added drop by drop until all but the carbon is dissolved. The bulk should not be allowed to exceed 60 c. c. The liquid is transferred to a 100 c. c. flask, 0.5 gram of dry calcium chlorid, a few drops of phenolphthalein solution added, then a 10 per cent. solution of sodium hydroxid until a permanent pink color is produced, and then 25 c. c. of lime-water. All the phosphate will be thus precipitated as calcium phosphate. The mixture is made up to 100 c. c., well mixed, and 50 c. c. collected through a dry filter. Normal sulfuric acid is added to this until the pink color is discharged, then methyl orange is added and the acid again dropped until the yellow tinge is just changed to pink. Fifth-normal sodium hydroxid is added (with care to avoid excess) until the liquid is yellow. At this stage all acids likely to be present exist as salts, neutral to phenolphthalein, except boric acid (which, being neutral to methyl orange, is in the free condition) and a small amount of carbonic acid that may be expelled by a few minutes' boiling. The solution is cooled, a little phenolphthalein added,

and as much glycerol as will give at least 30 per cent. of that substance in the solution, and then titrated with fifth-normal sodium hydroxid until the pink color appears. Each c. c. of the alkaline solution equals 0.0124 of crystallized boric acid. The process is satisfactory with milks containing not over 0.2 per cent. of boric acid. The charring must be carried only so far as to obtain a residue which will give a clear solution with the hydrochloric acid.

Boric acid may also be estimated by Gooch's method which is described in the analytic manuals. Hehner (*Analyst*, 1891) has modified the method by substituting sodium phosphate for powdered lime. (See page 52.)

Salicylic Acid.—Fifty c. c. of the sample are treated with acid mercuric nitrate for the removal of the fat and proteids, as described in connection with the determination of milk sugar, and the liquid filtered. The filtrate is shaken violently with about one-half its volume of a mixture of equal parts of ether and petroleum ether. The ethereal liquid is evaporated and a drop of neutral solution of ferric chlorid added to the residue. If salicylic acid be present, a characteristic violet color is developed. The reaction is very delicate.

Sodium Carbonate.—The following method, due to E. Schmidt, is stated to be capable of detecting one-tenth of one per cent. of sodium carbonate, or of sodium acid carbonate.

Ten c. c. of the milk are mixed with an equal volume of alcohol, and a few drops of a one per cent. solution of rosolic acid added. Pure milk shows merely a brownish-yellow color, but in the presence of sodium carbonate a

more or less marked rose-red appears. The delicacy of the test is enhanced by making a comparison cylinder with the same amount of milk known to be pure. If the salt is present in considerable amount, it may be detected by the increase in the ash, its marked alkalinity and effervescence with acid.

Preservation of milk samples.—For preserving milk samples for future analysis or comparison, various substances have been suggested, among which are hydrofluoric acid, alcohol, chloroform, carbon disulfid and formaldehyde. Richmond reported (*Analyst*, 1889, p. 2) good results by the addition of 0.5 per cent. of commercial hydrofluoric acid. In a discussion at a meeting of the Society of Public Analysts, in October, 1894 (*Analyst*, 1894, p. 247), Allen stated that he had added to each sample twice its weight of alcohol, but was not satisfied with this method, although it had, in some instances, served a good purpose. Mr. Richmond stated that he had been using formalin for a year and a half; 0.05 per cent. will keep milk for a month and larger quantities for an indefinite period.

Bevan has, however, noted the fact (*Analyst*, 1895, p. 152) that the total solids of milk containing formaldehyde are always higher, and that the increase is much greater than can be accounted for, even assuming that the whole of the formaldehyde remains in the residue, which is improbable, since it is volatile. Experiments on pure solutions of albumin, milk-sugar, and cane-sugar showed in each case an increase in residue when evaporated with formaldehyde. He has suggested that a small part, at least, of the increase in weight observed, is due to the formation of a non-volatile polymer of formaldehyde, and

in the case of milk-residues the increase is largely due to the conversion of milk-sugar into galactose.

Commercial solutions of formaldehyde, even after recent re-distillation, leave considerable residue,—probably the polymer, paraformaldehyde,—when evaporated in a flat dish in the manner employed in determining milk-solids.

Addendum to detection of boric acid.—E. A. Farrington (*J. A. C. S.*, Sept., 1896) has found that "preservaline" (which contains boric acid) added to fresh milk, increases the acidity four times as much as when added to water in the same proportion. He considers, therefore, that a milk which exhibits an acidity equal to 0.36 per cent. of lactic acid (8 c. c. of decinormal alkali to 20 c. c. of milk), and does not smell nor taste sour, has been probably adulterated with boric acid, since that proportion of lactic acid would be distinctly evident to the senses.

DATA FOR MILK INSPECTION.

VARIATIONS IN COMPOSITION.

Average Proportion of Solids in Milk.—The most extensive data on this point are those obtained by Vieth, a summary of whose results, covering a period of eleven years, was published in the *Analyst* of May, 1892. The total number of samples was 120,540, and although some changes had been made in the methods of analysis since the beginning of the work, the results were recalculated so as to be strictly comparable. The averages of the entire series were as follows :—

Fat, ...	4.1 per cent.
Non-fatty Solids, ...	8.8 "
Total Solids, ...	12.9 "

Seasonal Variations in the Composition of Milk.—The diagrammatic synopsis of Vieth's results (*Analyst*, 1892) shows that a notable variation in the proportion of ingredients occurs during the year. The poorest quality occurs during the first half of the year, especially in April. A low figure is also frequently noted about July. In autumn the quality rises, being highest in October and November.

The figures show that the variations in the total solids are due mainly to the variations in fat, but not entirely, for an increase in the proportion of fat is usually attended by a slight increase in the non-fatty solids.

The earlier tendency was to assign too high a limit for the non-fatty solids, since this figure was obtained by methods which failed to extract all the fat. In applying, therefore, the more modern processes, normal milk will be found to yield a figure for the non-fatty solids decidedly below the extreme limit of 9.5 per cent. Even nine per cent. for the non-fatty solids is more than is usually present.

While it may be permissible in special cases, such as the purchase of milk under contract, or in the operation of a large dairy, to reject samples which yield below nine per cent. of non-fatty solids, it is not just to exact such a standard for purposes of public inspection, and as a basis for penal proceedings. The standard of the Society of Public Analysts of England (8.5 per cent. of non-fatty solids) has been found satisfactory in the large experience of the members of that body, and Dr. Vieth has expressed himself as follows concerning it:

"My object is by no means to raise the cry that the standard adopted by the Society is too high; on the contrary, I think it is very judiciously fixed, but in upholding the standard of purity it should not be forgotten that the cows have never been asked for, nor given their assent to it, and that they will at times produce milk below standard. A bad season for hay-making is, in my experience, almost invariably followed by a particularly low depression in the quality of milk, toward the end of winter. Should the winter be of unusual severity and length, the depression will be still more marked. Long spells of cold and wet, as well as of heat and drought, during the time when cows are kept on pasture, also unfavorably influence the quality and, I may add, quantity of milk."

Deficient Solids.—The following are some instances of deficiency of solids in milk known to be genuine:—

Sp. Gr.	*Fat*	*S-N-F.*	*Total Solids*	*Analyst*
1029.6	3.38	7.95	11.33	C. B. Cochran
1030.0	3.62	8.31	11.93	"
1029.3	3.63	8.02	11.65	"
. . .	3.99	8.36	12.35	Leffmann and Beam
. . .	3.11	8.33	11.44	Monthly Averages
. . .	3.05	8.33	11.38	N. J. State Agric'l
. . .	3.23	8.44	11.67	Exp. Station

The following analyses have been kindly furnished us by C. B. Cochran, in advance of publication by him. The samples were taken under precautions which ensured their genuineness. The data are all direct determinations. The total solids were obtained by drying in the usual manner, and the fat by the L-B. method. Low milks have been often noted in the vicinity of Philadelphia.

Cow	*Sp. Gr.*	*Fat*	*S-N F.*	*Total Solids*
Pansy, . . .	1026.6	2.35	6.78	9.13
Rosie, . . .	1028.8	2.95	7.56	10.51
Dubell, . . .	1028.8	2.40	7.56	9.96
Gussie, . . .	1033.5	2.90	8.68	11.58

R. Bodmer (*Analyst*, 1895) reported the analysis of a sample obtained from one cow (June 13, 1895) as follows:

Sp. Gr.	*Fat*	*Proteids*	*Sugar*	*Ash*
1024.8	3.14	3.77	2.59	0.88

In a herd of 60 cows, Richmond found 19 per cent. of the samples to contain between 8.38 and 8.50 per cent. solids-not-fat.

The following instances of unusually rich milk, were reported in the *Analyst*, January, 1893:—

Sp. Gr.	*Total Solids*	*Fat*	*Ash*	*Analyst*
1026.6	19.50	11.06	.53	Smetham
1027.8	16.06	7.37	.72	"
1031.5	14.98	3.92	—	De Hailes

Since a partial creaming takes place in the udder, the first milkings (fore-milk) are poorer, and the last milkings (strippings) richer in fat, than the average milk. To insure a proper sample, the entire milking must be taken.

Babcock (Proc. 12th Annual Conv. A. O. A. C., p. 123) states that during the protracted drought of the summer of 1895, the average of nearly 100 determinations at the University of Wisconsin creamery gave but a trifle over 8.5 per cent. solids-not-fat. The casein was low in this milk, while the sugar was about normal in amount. Similar conditions have been observed by Van Slyke at the New York Station.

Variation According to Breed.—The following figures, taken from Bulletin 77 (1890), New Jersey State Agricultural Experiment Station, show the average composition of milk of various breeds of cattle:—

AVERAGE COMPOSITION OF MILK FOR EIGHT MONTHS.

Herd	Specific Gravity	Percentage					
		Water	Total Solids	Fat	Casein	Sugar	Ash
Ayrshire, . . .	1034 1	87.30	12.70	3.68	3.48	4.84	0.69
Guernsey, ...	1035.0	85.52	14.48	5.02	3.92	4.80	0.75
Holstein-Friesian,	1032.8	87.88	12.12	3.51	3.28	4.69	0.64
Jersey, . .	1035.3	85.66	14.34	4.78	3.96	4.85	0.75
Short-Horn, . .	1033.9	87.55	12.45	3.65	3.27	4.80	0.73

Variations According to Season.—The following table is condensed from the above report:—

	Ayrshire		Holstein-Friesian		Jersey		Guernsey		Sh't-Horn	
	Total Solids	Fat	Total Solids	Fat	Total Solids	Fat	Total Solids	Fat	Total Solids	Fat
March, . .	13.00	3.95	12.46	3.89	14.99	5 36	15.29	5.46	13.99	4.69
April, . . .	13.09	3.85	12.39	3.84	14.83	5.32	14.95	5.20	12.76	3.89
May, ...	12.97	3.54	12.57	3.65	13.67	4.30	14.00	4.57	12.05	3.24
June, ...	12.58	3.42	12.99	3.73	13.42	4.08	13.86	4.55	11.97	3.23
July, ...	12.72	3.71	11.44	3.11	13.46	4.13	13.85	4.54	11.89	3.28
August, . . .	13.08	4.07	11.38	3.05	13.60	4.22	13.93	4.81	12.08	3.56
September, .	11.85	3.26	11.67	3.23	15.00	5.08	14.67	5.22	12.24	3.47
October, . .	12.27	3.60	12.08	3.55	15.75	5.71	15.28	5.78	12.61	3.82

Milk Standards.—Many efforts have been made to establish a minimum for the composition of normal milk, with a view to prevent adulteration. Standards proposed some years back, requiring a high proportion of non-fatty solids, were based upon analyses by methods which fail to extract the whole of the fat from the milk residue. The Society of Public Analysts of England formerly used a standard of nine per cent. non-fatty solids and 2.5 per cent. fat, but when the improved method of analysis was adopted, altered the standard to the figures given in the table. The following are some of the standards which have been adopted:—

State, City, Etc.	Percentage by Weights of Solids		
	Non-fatty	Fat	Total
Pennsylvania, 1885,	9.50	3.00	12.50
New York, 1884,	9.00	3.00	12.00
New Jersey, 1882,	9.00	3.00	12.00
Massachusetts, 1886,	9.30	3.70	13.00 May and June 12.00
Minnesota, 1889,	9.50	3.50	13.00
Columbus, Ohio,	9.375	3.125	12.50
Baltimore, Md.,			12.00
Denver, Col.,			12.00
Lansing, Mich.,		3.00	12.50
Madison, Wis.,		3.00	
Burlington, Vt.,		3.50	12.50
Des Moines, Iowa,		3.50	13.13
Portland, Oregon,			12.00
Omaha, Nebraska,		3.00	12.00
U. S. Treasury Department,	9.50	3.50	13.00
Philadelphia, 1890, Ordinance,	8.50		12.00
Society of Public Analysts,	8.50	3.00	11.50

SANITARY RELATIONS.

Several practical points of comparatively recent development are intimately connected with the work of the milk-analyst and need some discussion. It is now well recognized that the dairy is an important factor in the distribution of disease, the influence taking place through several channels.

Dairy-cattle are subject to several virulent infectious diseases which are communicable to man. The most important of these is tuberculosis. The tendency at the present time is to the view that the keeping of dairy-cattle is a fruitful cause of the spread of this disease. The specific germ of tuberculosis may be conveyed both in meat and

milk, and since the infection of the animal is not always recognized promptly, a most insidious source of danger exists. Other infectious and dangerous diseases, *e g.*, scarlet fever, diphtheria, and typhoid fever, may be conveyed by milk.

The common methods of adulterating milk, namely, by abstracting fat or adding water, diminish the food-value, but there has been great exaggeration of the importance of these changes. It can scarcely be sound to declare, as has occasionally been done by those engaged in promoting sanitary legislation, that milk reduced in fat by legitimate processes, or even watered to a considerable extent, is unwholesome. It is occasionally stated that the digestion of the proteids of milk is dependent on the presence of a certain amount of fat, but the experimental or clinical evidence of this is, apparently, not precise. Several competent authorities, *e. g.*, Vieth, Uffelmann, and Hartshorne, have unhesitatingly declared even closely-skimmed milk to be wholesome. As regards watered milk, it would be preposterous to assert that an article which is wholesome when containing nine per cent. of non-fatty solids, becomes unwholesome when containing eight per cent.

Efforts have been made in some localities to interfere with the sale of closely-skimmed milk, especially that obtained by the centrifugal method. The low fat-content is the only point in which this differs appreciably in composition from ordinary milk. Since the proteids, sugar, and mineral matters are still in normal amount, skim-milk must have much food value. Questions of a broader character are, however, to be considered. Grotenfeldt ("Modern Dairy Practice," English Edition, trans. by F. W. Woll) discusses the matter at length and shows that the sepa-

rator skim-milk, as compared with that obtained by the setting process, is purer and fresher. It has fewer bacteria than the milk from which it was prepared, and has been freed from the disagreeable and disgusting adventitious matters that are so often present in whole milk. Concerning the objection to the low fat-content upon which is sometimes based a claim for a distinction between "skimmed milk" and "separator-milk," applying the former term to milk treated by the setting process, it must be noted that all but a very small amount of the fat may be removed by allowing the milk to stand for several days at a low temperature.

The unwholesomeness of milk arises not from change in the proportions of its principal ingredients, but from contamination with microorganisms. The danger from certain specific organisms has been mentioned, but the more frequent danger is from the ordinary non-pathogenic or putrefactive microbes, which, unless special care be taken, are invariably present and multiply rapidly. To prevent such conditions, resort is had to sterilization by heat. Brief exposure to a temperature of 100° C. is sufficient in most cases, but if the milk be subsequently exposed to air at ordinary temperatures, or mixed with unboiled water, it will be again contaminated and undergo putrefactive changes. In the warmer seasons of the year, these changes occur with great rapidity. Since clinical experience seems to show that boiled milk is frequently an unsatisfactory food for infants, methods of fractional sterilization at lower temperatures have been suggested. These depend on the fact, that, while spores and immature microbes require a rather high temperature for their destruction, fully developed organisms are more easily killed. By heating the

milk, therefore, to a temperature much below the boiling point, the adult microbes are killed, while the milk-solids are not unfavorably affected. The spores and immature organisms will, however, survive and may in a few hours develop; hence the milk is again heated, as before, and these later developed organisms will be killed. This process is repeated several times and finally complete sterilization is effected.

For the practical purpose of rendering milk safe as an article of food, it is not necessary to make repeated heatings. Numerous investigations are reported on this point, one of the more recent being a paper by Dr. R. G. Freeman (*Med. Rec.*, June 10, 1893). A temperature of 167° F. (75° C.) continued for fifteen minutes, followed by rapid cooling by immersing the containing vessel in water, will kill the adult forms of most microbes, and milk so treated will remain unaltered for one or two days and will not have suffered any appreciable loss of digestibility, even for infants.

When it is considered that milk is almost the only form of animal food that is eaten in the uncooked condition, by civilized communities, the importance of the facts above noted will be apparent. Some interesting data as to the association of consumption, diphtheria, and similar diseases with the maintenance of dairies have been collected, but the discussion of this feature of the question would be out of place here. Enough is known to show that raw milk is not a safe article of food, unless collected with such precautions as will prevent the introduction of infectious matter.

Artificial coloring matters do not involve any serious danger to health, except Martius' yellow (dinitroalpha-

naphthol) which is poisonous. The obvious objection to their use is that they enable milk of inferior quality to be substituted for rich milk. It is worthy of note that the assertion, occasionally made, that urine is employed in the preparation of annatto, is of little weight, since the annatto sold for dairy use is prepared by unobjectionable methods.

Abnormal Milks.—Milk occasionally becomes blue on the surface, the color forming in patches in proportion as the cream rises. The condition is due to the development of a chromogenic bacillus, first noted by Ehrenberg, and by him called *Vibrio syncyanus*, but now more correctly called *Bacillus syncyanus*. The condition sometimes prevails in epidemic form. The butter prepared from such milk possesses a greenish color, and a disagreeable butyric odor. The bacillus seems to be non-pathogenic. Hueppe fed animals on food mixed with strong cultures of it, and observed no serious results. To prevent the development and spread of the bacillus it is recommended that the vessels intended to receive the milk be washed in boiling water. Reiset states that blue milk may be used for the production of butter by adding 0.5 gram of acetic acid to each liter (eight grains to the quart).

Red milk is due to accidental contamination with the *Bacillus prodigiosus*. The spores of this microbe exist in the atmosphere and rapidly develop when they fall upon any nutritive medium. The microbe does not appear to have any pathogenic properties.

Ropy Milk.—This condition is occasionally seen dur-

ing moist warm weather. The milk when drawn may not show any unusual properties, but in a few hours becomes so viscid that a spoonful of it may be lifted several inches without breaking the connection between the two portions. The nature and cause of the change are not known. The phenomenon generally appears rather suddenly and does not last long, almost always disappearing promptly on the advent of colder weather. Cases are known in which the milk thus affected has been used as food without any apparent unfavorable effect.

MILK PRODUCTS.

CONDENSED MILK.

A few brands of condensed milk in the market under the name of "evaporated cream," consist merely of whole milk concentrated to about two-fifths of its bulk, but most condensed milks contain a considerable amount of cane-sugar. These samples represent, usually, whole milk concentrated to about one-third or two-sevenths of its original volume. A small amount of invert-sugar may be present. Portions of the lactose may crystallize from condensed milk, and when solutions are prepared for analysis, abnormal polarimetric reading will result unless the liquid stands a considerable time or is heated for a short time to 100° C.

The analysis of unsweetened condensed milks is conducted as with ordinary milk, the sample having been previously diluted with several times its weight of water heated to boiling, cooled and made up to a definite volume. The fat may be readily estimated in unsweetened milks by the L-B. process, but in sweetened ones, the charring action of the sulfuric acid seriously interferes. We find that mixing 5 c. c. of diluted condensed milk with 3 c. c. of the hydrochloric-fusel-oil mixture in the test-bottle, filling up to the neck with glacial acetic acid and heating for a few minutes in boiling water will avoid charring and allow the fat to be brought up by the centri-

fugal machine, but we have not tested the method sufficiently to ascertain its accuracy.

The most common defect in condensed milks is deficiency in fat, due to preparation from closely-skimmed milks. English analysts have frequently called attention to such samples, but the reports of analysts in the United States indicate that such practices are uncommon in this country. We have from time to time examined very cheap samples of condensed milk sold in Philadelphia, but have not found any marked deficiency in fat. Attention has been called, especially by Allen, to the fact that labels on condensed-milk tins often advise such large additions of water as to produce a weak milk. Preservatives (other than cane-sugar) and coloring matters are rarely if ever used, nor is it likely that foreign fats will be present.

The presence of cane-sugar, and possibly of invert-sugar, renders the complete analysis of condensed milk rather tedious, but for many practical purposes it will be sufficient to determine the total solids, fat, and proteids. If the ash and milk-sugar also be determined, the cane-sugar may be approximately estimated by difference. In all examinations of condensed milk, care should be taken that the contents of the can are thoroughly mixed before any sample is taken. The following procedures cover the usual analysis of ordinary samples, and are partly those given by Pearmain and Moor.

Ten grams of the sample should be dissolved in water and made up to 100 c. c. Aliquot portions of this solution should be taken for analysis.

Total Solids.—Five to ten c. c. are evaporated by the Babcock method (p. 21), or on ignited asbestos in a shallow platinum basin.

Ash.—Twenty c. c. or more are evaporated in a platinum dish and the ash determined with the usual precautions.

Fat.—The cylinder containing the residue from the determination of total solids is placed in the extractor and the fat obtained in the usual way. Another method is to distribute five c. c. over fat-free paper, proceeding as in the original Adams' process. The Werner-Schmidt method is not suitable.

Proteids.—These determinations are made as with ordinary milk.

Sugars.—If regard is to be had to the possible presence of invert-sugar, a special method must be followed which is described below. The processes first given consider lactose and sucrose only.

Lactose.—Richmond and Bosely have shown that heating to the extent to which milk is subjected in the preparation of condensed milk may reduce the rotatory power of the sugar sufficiently to cause serious error, if the polarimeter be used for the determination. The reducing power with alkaline copper solutions is not seriously affected.

Sucrose.—The determination of cane-sugar may be made by difference, that is subtracting the sum of the other ingredients from the total solids. Such a method is, of course, approximate only, but it may serve for ordinary inspection purposes, since the amount present is almost always large, generally more than the milk-solids themselves, and an error even of several per cent. does not affect the judgment as to the wholesomeness of the sample. More exact work requires, however, that the cane-sugar shall be determined directly, and several processes have been devised for the purpose. Sucrose exerts but little action

on Fehling's solution, but invert-sugar acts powerfully, and one set of processes depends on determining the reducing power before and after inversion. Since the polarimetric reading is also markedly changed by the inversion, the difference in polarization may be employed. Processes of fermentation may be so conducted as to remove the sucrose (also any form of glucose) while the lactose is unaffected. This method is chiefly valuable for recognizing invert-sugar or either of its constituents.

When inversion methods are used, they must be such as to secure prompt inversion of the sucrose without affecting the lactose. Experiment shows that citric acid and invertase (the enzyme of yeast) are the most suitable agents. Stokes and Bodmer (*Analyst*, 1885, p. 62) have worked out the citric acid method substantially as follows:—

About eight grams of the sample are accurately weighed, diluted with water, coagulated with about one per cent. of citric acid, without heating, and made up to 200 c. c. plus the volume of the precipitated fat and proteids (see p. 39). The liquid portion, which now measures 200 c. c., is passed through a dry filter. The reducing power with alkaline copper solutions is determined at once upon 50 c. c. of this filtrate. To another 50 c. c., one per cent. of citric acid is added and the solution boiled for ten minutes and the reducing power also determined. The increase over that of the first solution is due to the invert-sugar formed by the action of the citric acid on the sucrose. It is necessary to bear in mind that the reducing equivalents of lactose and invert-sugar are not the same. Stokes and Bodmer prefer to work with Pavy's solution. The following description is partly from their paper and partly from Allen's "Chemistry of Urine."

Pavy's solution :—

Pure crystallized copper sulfate, .	. 4.157	grams
Pure crystallized Rochelle salt, . .	21.6	grams
Sodium hydroxid of good quality, . .	20.5	grams

The copper sulfate is dissolved in about 100 c. c. of hot water, the Rochelle salt and sodium hydroxid are dissolved in another portion of warm water, the solutions allowed to cool and mixed, 400 c. c. of ammonium hydroxid (sp. gr. .880) added and the liquid made up to one liter.

Of this solution,

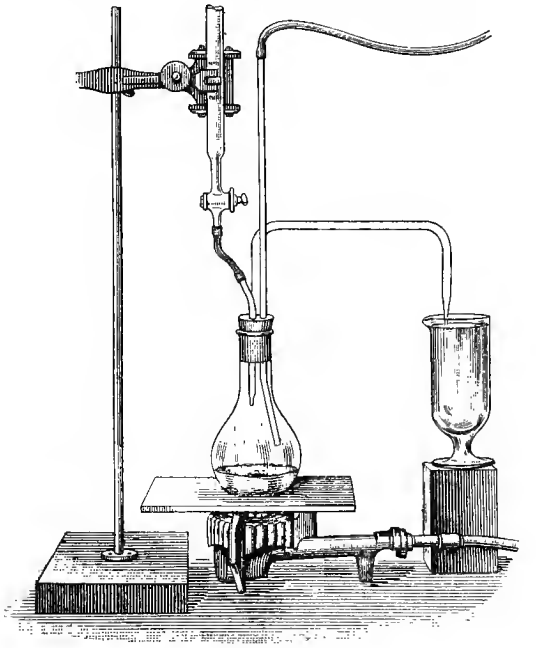
10 c. c. equals 0.005 dextrose
10 c. c. equals 0.0096 lactose
10 c. c. equals 0.0047 sucrose (after inversion).

The best results are obtained only when working so as to exclude free access of air. Allen suggests the following arrangement : To the lower end of the buret is attached a short rubber tube which connects with a glass tube that passes through a rubber stopper fitted in a flask of convenient size. The stopper has two other openings, one for the admission of a tube through which a slow current of illuminating gas or hydrogen may be transmitted in order to maintain a non-oxidizing atmosphere ; the other for a tube for the escape of steam and ammonium hydroxid.

The gas connection may be omitted if, as suggested by Stokes, the tubes through which the vapors escape be joined with an empty Woolff's bottle connected by a tube to a vessel containing cold water. The flask containing the copper solution may be placed upon a whitewashed iron plate in order to show the tint of the solution more clearly.

From 25 to 40 c. c. of the copper solution, accurately measured, are placed in the flask, a few fragments of pumice dropped in, the tubes and buret adjusted, and the solu-

tion brought to boiling. The liquid containing the lactose is added in small portions. Since the oxidizing action occurs more slowly than with glucose, the additions must be at greater intervals. The process is finished as soon as the



From Allen's "Chemistry of Urine."

liquid is colorless. When common coal gas flows into the flask, a brick-red film of cuprous acetylid is formed on the surface of the liquid from the acetylene present in the gas.

It is necessary to verify the correctness of the copper

solution and this may be done by means of known weights of pure lactose and sucrose, the latter being first inverted by the citric acid as described.

Allen has recently (*Analyst*, 1895, p. 127) stated that the Gerrard-Allen method—titration with cupric cyanid—promises to be the most satisfactory volumetric method for glucose and that it may answer also for other sugars. It is described in full in *Pharmaceutical Journal* for April 20, 1895, also in the "Chem. of Urine," above cited, p. 74.

The following method is based on the difference in polarimetric reading before and after action of invertase. About 30 grams of the sample are accurately weighed in a 100 c. c. flask, diluted to about 80 c. c., heated to boiling, cooled, and 7.5 c. c. of acid mercuric nitrate solution added. The mixture is made up to 100 c. c., well shaken, filtered through a dry filter and the polarimetric reading taken at once. It will be the sum of the effect of the two sugars. The volume of the sugar-containing liquid is calculated by allowing for the precipitated proteids and fat, as described on p. 39.

Fifty c. c. of the filtrate are placed in a flask marked at 55 c. c., a piece of litmus paper dropped in, and the excess of nitric acid cautiously neutralized by sodium hydroxid solution. The liquid is then faintly acidified by a single drop of acetic acid (it must not be alkaline), a few drops of an alcoholic solution of thymol added, and then 2 c. c. of a solution of invertase, prepared by grinding half a cake of ordinary compressed yeast with 10 c. c. of water and filtering. The flask is corked and allowed to remain at a temperature of 35° to 40° C. for twenty-four hours. The cane-sugar will be inverted while the milk-sugar will

be unaffected. The flask is filled to the mark (55 c. c.) with washed aluminum hydroxid and water, mixed, filtered, and the polarimetric reading taken.

A powerful solution of invertase may be prepared by the method recommended by O'Sullivan and Tompson. Brewer's yeast is allowed to stand at a temperature of 15° C. for a month. The liquid is filtered and sufficient alcohol added to give about 12 per cent. of absolute alcohol. After a few days the liquid is filtered and is ready for use. The alcohol acts as a preservative.

The rotatory powers of cane-sugar and dextrose are not appreciably affected by temperature within the limits of ordinary experiments. The same may be said of milk-sugar (see p. 41). Invert-sugar, by reason of the levulose present, is materially affected by the temperature. Thus, a solution of cane-sugar, which, before inversion, causes a rotation of + 100 angular degrees, has after inversion, if observed at 0° C., a rotation of —44 degrees, a total change of 144; but at 21° C. the reading will be only —33 angular degrees, a total change of 133. The following formula is to be used for calculation:—

$$C = \frac{100\ D}{144 - \frac{t}{2}}$$

in which C equals the angular rotation due to the uninverted cane-sugar, D the difference in the polarimetric reading before and after inversion, and t the temperature in Centigrade degrees. Since in the performance of the inversion, the liquid has been diluted from 50 to 55 c. c., the polarimetric reading must be increased in proportion, before the value of D is found.

The specific rotatory power of cane-sugar varies slightly with the concentration. Tollens gives the following formula, in which S is the specific rotatory power and C the concentration in grams per 100 c. c. :—

$$S = 66.386 + .015035\ C - .0003986\ C^2$$

Bigelow and McElroy (*J. A. C. S.*, Dec., 1893) propose the following routine method for the determination of the sugars, including invert-sugar, in condensed milk. The solutions used are :—

Acid mercuric iodid.—Mercuric chlorid, 13.5 grams; potassium iodid, 33.2 grams; glacial acetic acid, 20 c.c. ; water 640 c.c.

Alumina-cream.—A cold saturated aqueous solution of alum is divided into two unequal portions; to the larger portion ammonium hydroxid is added to slight excess, and then small portions of the remaining solution until a slight acid reaction is secured.

The entire contents of the can are transferred to a porcelain dish and thoroughly mixed. A number of portions of about 25 grams are weighed carefully in 100 c.c. flasks. Water is added to two of the portions, and the solutions boiled. The flasks are then cooled, clarified by means of a small amount of the acid mercuric iodid and alumina-cream, made up to mark, filtered and the polarimetric reading noted. Other portions of the milk are heated in the water-bath to 55° C.; one-half of a cake of compressed yeast is added to each flask and the temperature maintained at 55° C. for five hours. Acid mercuric iodid and alumina-cream are then added, the solution cooled to room-temperature, made up to mark, mixed, filtered, polarized. The amount of cane-sugar is determined by

formula on page 71. Correction for the volume of precipitated solids may be made by the double dilution method (p. 41). The total reducing sugar is estimated in one of the portions by one of the reducing methods, and if the sum of it and the amount of cane-sugar obtained by inversion is equal to that obtained by the direct reading of both sugars before inversion, no invert-sugar is present. If the amount of reducing sugar seems to be too great, the milk-sugar must be re-determined as follows: Two hundred and fifty grams of the condensed milk are dissolved in water, the solution boiled, cooled to 80° C., a solution of about four grams of glacial phosphoric acid added, the mixture kept at 80° C. for a few minutes, then cooled to room temperature, made up to mark, shaken and filtered. It may be assumed that the volume of the precipitate is equal to that obtained by mercuric iodid solution. Enough potassium hydroxid is then added to not quite neutralize the free acid, and sufficient water to make up for the volume of the solids precipitated by the phosphoric acid. The mixture is then filtered and the filtrate is measured in portion of 100 c.c. into 200 c.c. flasks. A solution containing 20 milligrams of potassium fluorid and half a cake of compressed yeast is added to each flask, and the mixture allowed to stand for ten days at a temperature of from 25° C. to 30° C. The invert-sugar and cane-sugar are fermented and removed by the yeast in the presence of a fluorid, while milk-sugar is unaffected. The flasks are filled to the mark and the milk-sugar determined either by reduction or by the polariscope. The amount of copper solution reduced by the lactose and invert-sugar, less the equivalent of lactose remaining after fermentation, is due to invert-sugar.

TABLE OF ANALYSES OF CONDENSED MILKS.

Brand.	Total Solids.	Fat.	Proteids	Lactose.	Sucrose.	Ash.	Analyst.
First Swiss, .	36.70	10.5	9.7	14.2	none	2.1	Pearmain and Moor.
Highland, .	31.25	9.63	9.21	10.89	none	1.52	F. T. Aschman.
Howell's, .	28.08	8.81	8.53	9.82	none	1.82	" "
St. Charles, .	33.54	9.26	10.49	12.24	none	1.55	" "
Anglo-Swiss,	78.44	9.37	9.16	13.39	40.45	2.07	" "
Dime, .	76.12	7.34	10.07	12.70	43.95	1.96	" "
Eagle, .	69.84	7.51	8.40	9.82	42.24	1.87	"
Milk-maid,	74.24	9.03	9.33	10.18	43.72	1.98	" "
Nestlé, .	75.80	9.81	10.49	11.66	41.63	2.21	" "
Calf, .	58.00	1.0	7.5	16.0	31.9 (by diff.)	1.6	Pearmain and Moor.
Cowslip,	70.90	1.4	11.4	14 6	41.9 (by diff.)	1.6	" "

BUTTER.

Butter, commercially, consists of a variable mixture of fat, water, and curd, obtained by churning cream from cow's milk. The water contains in solution milk-sugar and the salts of the milk. Common salt is usually present, being added after the churning. Artificial coloring is frequently used.

The composition of commercial butter usually varies within the following limits :—

Fat,	. . . . .	78	per cent.	to	94	per cent.
Curd,	. . . . .	1	" "	"	3	" "
Water,	. . . . .	5	" "	"	14	" "
Salt,	. . . .	0	" "	"	7	" "

Nostrums for Butter Making.—Preparations are sold purporting to have the power to increase the yield of butter from a given weight of milk. One of these, advertised under the name "black pepsin," has been found to contain salt, annatto, and a small amount of rennet. Pepsin has also been used. These curdle the milk and allow the incorporation of much cheese and water with the butter. Butter may also, without the addition of chemicals, be incorporated with a large amount of cream.

We have encountered butter containing over forty per cent. of water. Such samples are pale and spongy, lose weight, and become rancid very rapidly.

It is generally considered that butter should not contain more than 16 per cent. water. An excess of water diminishes the keeping quality.

The following methods for the analysis of butter have been adopted by the A. O. A. C. :—

Sampling.—If large quantities of butter are to be sampled, a butter trier or sampler may be used. The portions drawn, about 500 grams, are to be carefully melted in a closed vessel, at as low a heat as possible, and when melted the whole is to be shaken violently for some minutes till homogeneous. The mass must be sufficiently solidified to prevent the separation of the water and fat. A portion is then placed in the vessel from which it is to be weighed for analysis, and should nearly or quite fill it. It should be kept in a cold place until analyzed. Determinations are made as follows :—

Water.—1.5–2.5 grams are dried to constant weight at the temperature of boiling water in a dish with a flat bottom, having a surface of at least 20 sq. cm. The use of clean, dry sand is admissible.

Fat.—The dry butter from the water determination is dissolved in the dish with absolute ether, or with 76° petroleum spirit. The contents of the dish are then transferred to a Gooch crucible with the aid of a wash-bottle filled with the solvent, and are washed until free from fat. The crucible and contents are dried at the temperature of boiling water, until the weight is constant.

Indirect Method.—Water may be determined by drying on asbestos or sand, and the fat extracted by ether.

Casein and Ash.—The crucible containing the residue from the fat determination, consisting of the casein and salts, is covered and heated, gently at first, and gradually raising the temperature to just below redness. The cover may then be removed and the heat continued till the contents of the crucible are white. The loss in weight of the crucible and contents represents the weight of the casein, and the residue in the crucible, ash. The mineral matter may be dissolved in water very slightly acidulated with nitric acid, and the chlorin determined in the usual way.

Salt.—Weigh in a counterpoised beaker from 5 to 10 grams of the sample. The butter is placed, in portions

of about 1 gram at a time, in the beaker, these portions being taken from different parts of the sample. Hot water is now added (about 20 c. c.) to the beaker containing the butter, and after it has melted the liquid is poured into the bulb of a separating funnel. The stopper is now inserted and the contents shaken for a few moments. After standing until the fat has all collected on top of the water, the stopcock is opened and the water is allowed to run into an Erlenmeyer flask, being careful to let none of the fat globules pass. Hot water is again added to the beaker, and the foregoing process is repeated from ten to fifteen times, using each time 10 to 20 c c. of water. The resulting washings contain all but a mere trace of the NaCl originally present in the butter. The sodium chlorid may be determined volumetrically in the filtrate.

Antiseptic substances in milk may find their way into the butter made from it. They will be dissolved in the water, and may be detected by separating this, by melting, and testing it as directed under milk.

OLEOMARGARIN.—Under this term is now included by act of Congress, any oleaginous substance intended as a substitute for butter, containing any proportion of fat other than butter-fat. The term "margarine" is employed in England, under authority of an act of Parliament, with the same significance. The principal materials employed in the preparation of butter substitutes are cottonseed oil, mutton-fat, and beef-fat.

When fats are saponified and the soap treated with acid, the individual fatty acids are obtained. It is upon the recognition of the peculiar acid radicles existing in butter that the most satisfactory method of distinguishing it from other fats is based. Since the relative proportion of these radicles differs in different samples, the quantitive estimation cannot be made with accuracy, but when the foreign

fats are substituted to the extent of 25 per cent. or more, the adulteration can be detected with certainty and an approximate quantitive determination made.

Distillation method.—The fatty acids containing a small number of carbon atoms, set free by the process noted above, are soluble in water and volatile. A method for their estimation depending on their solubility in water was perfected by Hehner, but has now been displaced by a distillation method originally suggested by Hehner & Angell, but improved by Reichert, and the details perfected by others, especially Wollny, and now generally known as the Reichert-Wollny method.

We have modified the process by substituting a solution of sodium hydroxid in glycerol as the saponifying agent, by which the time required is much shortened, the result subject to less variation, and the titration more exact. The following reagents are required.

Glycerol-soda.—One hundred grams of pure sodium hydroxid are dissolved in 100 c. c. of distilled water, and allowed to stand until clear. Twenty c. c. of this solution are mixed with 180 c. c. of pure concentrated glycerol. The mixture can be conveniently kept in a capped bottle holding a 10 c. c. pipet, with a wide outlet.

Sulfuric acid.—Twenty c. c. of pure concentrated sulfuric acid, made up with distilled water to 100 c. c.

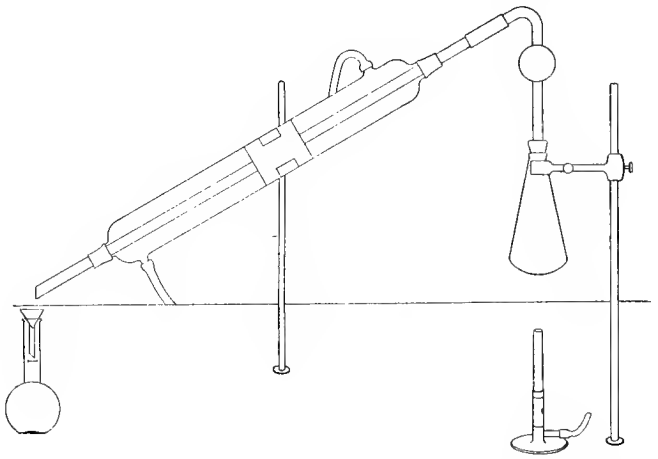
Barium hydroxid.—An approximately decinormal, accurately standardized, solution of barium hydroxid.

Indicator.—An alcoholic solution of phenolphthalein.

About 50 grams of the sample are placed in a beaker, and heated to a temperature of 50° to 60° C., until the water and curd have settled to the bottom. The clear fat is then poured on a warm, dry plaited filter, and kept in a

warm place until 25 or 30 c. c. have been collected. If the filtrate is not perfectly clear, it should be reheated for a short time and again filtered.

A 300 c. c. flask is washed thoroughly, rinsed with alcohol and then with ether, and thoroughly dried by heating in the water oven. After cooling it is allowed to stand for about fifteen minutes and weighed. A pipet, gradu-



ated to 5.75 c. c., is heated to about 60° C. and filled to the mark with the well-mixed fat, which is then run into the flask. After standing for about fifteen minutes the flask and contents are weighed. Twenty c. c. of the glycerol-soda are added and the flask heated over the Bunsen burner. The mixture may foam somewhat; this may be controlled, and the operation hastened by shaking the flask. When all the water has been driven off, the liquid

will cease to boil, and if the heat and agitation be continued for a few moments, complete saponification will be effected, the mixture becoming perfectly clear. The whole operation, exclusive of weighing the fat, requires less than five minutes. The flask is then withdrawn from the heat and the soap dissolved in 135 c. c. of water. The first portions of water should be added drop by drop, and the flask shaken between each addition in order to avoid foaming. When the soap is dissolved, 5 c. c. of the dilute sulfuric acid are added, a piece of pumice dropped in, and the liquid distilled until 110 c. c. have been collected. The condensing tube should be of glass, and the distillation conducted at such a rate that the above amount of distillate is collected in thirty minutes.

The distillate is usually clear; if not, it should be thoroughly mixed, filtered through a dry filter, and 100 c. c. of the filtrate taken. To the distillate about 0.5 c. c. of the phenolphthalein solution are added, and the standard barium hydroxid run in from a buret until a red color is produced. If only 100 c. c. of the distillate have been used for the titration, the number of cubic centimeters of barium hydroxid should be increased by one-tenth.

When it is intended merely to distinguish butter from oleomargarin, it will be sufficient to measure into the flask 3 or 6 c. c. of the clear fat, and operate upon this directly.

A blank experiment should be made to determine the amount of decinormal alkali required by the materials employed. With a good quality of glycerol, this will not exceed 0.5 c. c.

Butter (5 grams) yields a distillate requiring from 24 to 34 c. c. of decinormal alkali. Several instances have been published in which genuine butter has given a figure as low

as 22.5 c. c., but such results are uncommon. The materials employed in the preparation of oleomargarin yield a distillate requiring less than 1 c. c. of alkali. Commercial oleomargarin is usually churned with milk in order to secure a butter flavor, and thus acquiring a small amount of butter-fat yields distillates capable of neutralizing from 1 to 2 c. c. of alkali.

That more uniform results may be obtained with the glycerol-soda method than with the use of alcohol has been clearly shown. W. Karsch, assistant in the Dairy Institute at Hameln, Germany (*Chemiker-Zeitung*, 1896, 62), has subjected the method to careful comparative examination and finds it to be more satisfactory than the Reichert-Wollny method. Dr. Vieth, Director of the Institute, has employed it for several years in the analytic work of the station.

The method of determining the volatile acids of butter-fat, adopted by the A. O. A. C., is as follows :—

Sodium hydroxid solution.—One hundred grams of sodium hydroxid are dissolved in 100 c. c. of pure water. It should be as free as possible from carbonates, and be preserved from contact with the air.

Alcohol, of about 95 per cent., redistilled over sodium hydroxid.

Acid.—Solution of sulfuric acid containing 25 c. c. of strongest acid in 1000 c. c. of water.

Barium hydroxid.—An accurately standardized, approximately decinormal solution of barium hydroxid.

Indicator.—One gram of phenolphthalein in 100 c. c. of alcohol.

Saponification flasks, 250 to 300 c. c. capacity, of hard, well-annealed glass, capable of resisting the tension of alcohol vapor at 100° C.

A pipet, graduated to deliver 40 c. c.

Distilling apparatus.

Buret.—An accurately calibrated buret, reading to tenths of a c. c.

The butter or fat to be examined should be melted, and kept in a dry, warm place at about 60° C. for two or three hours, until the water and curd have entirely settled out. The clear supernatant fat is poured off and filtered through dry filter paper, in a jacketed funnel containing boiling water. Should the filtered fat in a fused state not be perfectly clear, the treatment above mentioned must be repeated.

The saponification flasks are prepared by having them thoroughly washed with water, alcohol, and ether, wiped perfectly dry on the outside, and heated for one hour at the temperature of boiling water. The flasks should then be placed in a tray by the side of the balance, and covered with a silk handkerchief until they are perfectly cool. They must not be wiped with a silk handkerchief within fifteen or twenty minutes of the time they are weighed. The weight of the flasks having been accurately determined, they are charged with the melted fat in the following way:—

A pipet with a long stem, marked to deliver 5.75 c. c., is warmed to a temperature of about 50° C. The fat having been poured back and forth once or twice into a dry beaker in order to thoroughly mix it, is taken up in the pipet and the nozzle of the pipet carried to near the bottom of the flask, having been previously wiped to remove any adhering fat, and 5.75 c. c. of fat are allowed to flow into the flask. After the flasks have been charged in this way, they should be re-covered with a silk handkerchief and allowed to stand fifteen or twenty minutes, when they are again weighed.

Ten c. c. of 95 per cent. alcohol are added to the fat in the flask, and then 2 c. c. of the sodium hydroxid solution; a soft cork stopper is now inserted in the flask and tied down with a piece of twine. The saponification is then completed by placing the flask upon the water or steam

bath. The flask, during the saponification, which should last one hour, should be gently rotated from time to time, being careful not to project the soap for any distance up its sides. At the end of an hour the flask, after having been cooled to near the room temperature, is opened. The stoppers having been laid loosely in the mouth of the flask, the alcohol is removed by dipping the flask into a steam bath. The steam should cover the whole of the flask except the neck. After the alcohol is nearly removed, frothing may be noticed in the soap, and to avoid any loss from this cause or any creeping of the soap up the sides of the flask, it should be removed from the bath and shaken to and fro until the frothing disappears. The last traces of alcohol vapor may be removed from the flask by waving it briskly, mouth down, to and fro. After the removal of the alcohol the soap should be dissolved by adding 100 c. c. of recently boiled distilled water, warming on the steam bath with occasional shaking, until solution of the soap is complete. When the soap solution has cooled to about 65° C., the fatty acids are separated by adding 40 c. c. of the dilute sulfuric acid solution mentioned above. The flask should now be re-stoppered as in the first instance, and the fatty acid emulsion melted by replacing the flask on the steam bath. According to the nature of the fat examined, the time required for the fusion of the fatty acid emulsions may vary from a few minutes to several hours.

After the fatty acids are completely melted, which can be determined by their forming a transparent oily layer on the surface of the water, the flask is cooled to room temperature, and a few pieces of pumice stone added. The pumice stone is prepared by throwing it, at a white heat, into distilled water, and keeping it under water until used. The flask is now connected with a glass condenser, slowly heated with a naked flame, until ebullition begins, and then the distillation continued by regulating the flame in such a way as to collect 110 c. c. of the distillate in, as nearly as possible, thirty minutes. The distillate should be received in a flask accurately graduated at 110 c. c.

The 110 c. c. of distillate, after thorough mixing, are filtered through dry filter paper and collected in a flask marked at 100 c.c. One hundred c.c. of the filtered distillate are poured into a beaker holding from 200 to 250 c.c., 0.5 c.c. of phenolphthalein solution added, and decinormal barium hydroxid run in until a red color is produced. The contents of the beaker are then returned to the measuring flask to remove any acid remaining therein, poured again into the beaker, and the titration continued until the red color produced remains apparently unchanged for two or three minutes. The number of cubic centimeters of decinormal barium hydroxid required should be increased by one-tenth.

Many other methods of detecting butter adulteration have been proposed. The distinction between butter and its substitutes is not so sharp as with the distillation method, but all are of more or less value and may aid in the detection of adulteration in doubtful cases.

Acetic acid reaction (Valenta's test).—This depends upon the behavior of butter and acetic acid, and is one of the most valuable of the simple tests. The strength of acid used may vary within certain limits, but it must always be standardized against a sample of pure butter-fat.

Three c. c. of the melted fat are placed in a dry test-tube, an equal volume of acetic acid added, and the mixture heated until solution has taken place. It is then allowed to cool spontaneously, and the temperature at which the liquid begins to be turbid noted.

Messrs. Chattaway, Pearmain, and Moor, as the result of a rather lengthy trial, prefer an acid of about 95.5 per cent. With a weaker acid the test is less sensitive. They call attention to the fact that the presence of moisture in the fat is one of the most fruitful sources of error, and recom-

mend filtration of the sample through dry filter paper before performing the test. They note further that undue heating of the sample, either at the time the test is being made, or previously, renders the determination unreliable. With the acid employed by them, the following figures were obtained expressed in Centigrade degrees :—

Butter-fat (24 samples)—	
Highest,	39°
Lowest,	29°
Mean,	36°
Oleomargarin (5 samples)—	
Highest,	97°
Lowest,	94°
Mean,	95°
Cottonseed oil, various samples,	71°, 75°, 71°, 85°, 86°, 88°, 89°
Peanut oil,	72°, 73°
Lard oil,	75°, 76°, 75°
Lard,	98°, 97°, 98°, 97°
Beef stearin,	100°
Lard stearin,	100°.

E. W. T. Jones (*Analyst*, 1894, p. 151) recommends the employment of a standard butter with which to standardize each fresh batch of acid, and dilution of the acid to such a point, that the turbidity temperature with this butter-fat is 60° C. In this way the results are comparable with those of previous tests.

Oleomargarin gives temperatures from 95° to 106° C., and generally from 100° to 102° C.

Iodin number.—The common oils and fats are mixtures of ethers of the acetic and oleic series. The former are saturated and therefore form only substitution compounds,

but the latter readily form additive compounds with the members of the chlorin group, and by estimating the amount of the element taken up under definite conditions, a measure of the amount of unsaturated radicles present is obtained.

Hübl's method with iodin is now employed. The A. O. A. C. process is as follows:—

(1) *Iodin solution.*—Dissolve 25 grams of pure iodin in 500 c. c. of 95 per cent. alcohol. Dissolve 30 grams of mercuric chlorid in 500 c.c. of 95 per cent. alcohol. The latter solution, if necessary, is filtered, and then the two solutions mixed. The mixed solution should be allowed to stand twelve hours before using.

(2) *Decinormal sodium thiosulfate solution.*—Take 24.6 grams of chemically pure sodium thiosulfate freshly pulverized as finely as possible and dried between filter or blotting paper. Make this up to 1000 c. c. at the temperature at which the titrations are to be made.

(3) *Starch paste.*—One gram of starch is boiled in 200 c. c. of distilled water for ten minutes, and cooled to room temperature.

(4) *Solution of potassium iodid.*—One hundred and fifty grams of potassium iodid are dissolved in water and made up to 1 liter.

(5) *Solution of potassium dichromate.*—Dissolve 3.874 grams of chemically pure potassium dichromate in distilled water, and make the volume up to 1 liter at the temperature at which the titrations are to be made.

Run 20 c. c. of the potassium dichromate solution, to which has been added 10 c. c. of the solution of potassium iodid, into a glass stoppered flask. Add to this 5 c. c. of strong hydrochloric acid. Allow the solution of sodium thiosulfate to flow slowly into the flask until the yellow color of the liquid has almost disappeared. Add a few drops of the starch-paste, and with constant shaking continue to add the sodium thiosulfate solution until the blue color just disappears. The number of cubic centimeters

of thiosulfate solution used, multiplied by 5, is equivalent to 1 gram of iodin.

About 1 gram of butter-fat is to be weighed in a glass-stoppered flask holding about 300 c. c., with the precautions mentioned for weighing the fat for determining volatile acids. The fat in the flask is dissolved in 10 c. c. of chloroform. After complete solution has taken place, 30 c. c. of the iodin solution are added. The flask is now placed in a dark place and allowed to stand, with occasional shaking, for three hours.

One hundred c. c. of distilled water are added to the contents of the flask, together with 20 c. c. of the potassium iodid solution. Any iodin which may be noticed upon the stopper of the flask should be washed back into the flask with the potassium iodid solution. The excess of iodin is now taken up with the sodium thiosulfate solution, which is run in gradually, with constant shaking, until the yellow color of the solution has almost disappeared. A few drops of starch-paste are then added, and the titration continued until the blue color has entirely disappeared. Toward the end of the reaction the flask should be stoppered and violently shaken, so that any iodin remaining in solution in the chloroform may be taken up by the potassium iodid solution in the water. A sufficient quantity of sodium thiosulfate solution should be added to prevent a reappearance of any blue color in the flask for five minutes.

At the time of adding the iodin solution to the fats, two flasks of the same size as those used for the determination should be employed for conducting the operation described above, but without the presence of any fat. In every other respect the performance of the blank experiments should be just as described. These blank experiments must be made each time the iodin solution is used.

Example—Blank determinations.

(1) 30 c. c. iodin solution required 46.4 c. c. sodium thiosulfate solution.

(2) 30 c. c. iodin solution required 46.8 c. c. of sodium thiosulfate solution.

Mean 46.6.

Per cent. of iodin absorbed.

Weight of fat taken, ...	1.0479	grams
Quantity of iodin solution used, . .	30.0	c. c.
Thiosulfate equivalent to iodin used, . . .	46.6	"
Thiosulfate equivalent to remaining iodin, .	14.7	"
Thiosulfate equivalent to iodin absorbed, .	31.9	"

Per cent. of iodin absorbed, $31.9 \times 0.0124 \times 100 \div 1.0479 = 37.75$.

The iodin number for butter may range from 20 to 38; for oleomargarin from 40 to 55; the method is of little use, therefore, in this connection.

Soluble and insoluble acids.—This method, originated by Hehner and Angell, has been improved by various chemists. Butter usually yields at least five per cent. of soluble and 89.5 of insoluble acids, but numbers slightly below these limits have been obtained in butters known to be pure. The following formula has been suggested for calculation of the amount of adulteration. F is the percentage of foreign fat and I the percentage of insoluble acid:

$$F = 13.3\ (I - 88).$$

The following are the reagents and manipulations prescribed by the A. O. A. C.:—

Decinormal sodium hydroxid.

Alcoholic potassa.—Dissolve 40 grams of good potassium hydroxid, free from carbonates, in one liter of 95 per cent. redistilled alcohol. The solution must be clear.

Semi-normal hydrochloric acid accurately standardized.

Indicator.—One gm. of phenolphthalein in 100 c. c. of alcohol.

About 5 grams of the sample are weighed into a saponification flask (p. 81), 50 c. c. of the alcoholic potassa solution added, and the flask stoppered and placed in the steam bath until the fat is entirely saponified. The operation may be facilitated by occasional agitation. The alcoholic solution is always measured with the same pipet, and uniformity further secured by allowing it to drain the same length of time (thirty seconds). Two or three blank experiments are conducted at the same time. In from five to thirty minutes, according to the nature of the fat, the liquid will appear perfectly homogeneous, saponification is complete, and the flask is removed and cooled. When sufficiently cool, the stopper is removed and the contents of the flask rinsed with a little 95 per cent. alcohol into an Erlenmeyer flask of about 200 c. c. capacity, which is placed on the steam bath, together with the blanks, until the alcohol is evaporated. Titrate the blanks with seminormal hydrochloric acid. Then run into each of the flasks containing the fatty acids 1 c. c. more of the hydrochloric acid than is required to neutralize the alkali in the blanks. The flask is then connected with a condensing tube, three feet long, made of small glass tubing, heated on the steam bath until the separated fatty acids form a clear stratum. The flask and contents are then cooled in ice-water.

The fatty acids having quite solidified, the liquid contents of the flask are poured through a dry filter into a liter flask, care being taken not to break the cake.

Between 200 and 300 c. c. of water are next brought into the flask, the cork with its condenser-tube reinserted, and the flask heated on the steam bath until the cake of acids is thoroughly melted. During the melting of the cake of fatty acids, the flask should occasionally be agitated with a revolving motion, but so that its contents are not made to touch the cork. When the fatty acids have again separated into an oily layer, the flask and its contents are cooled in ice-water and the liquid filtered through the same filter into the same

liter flask. This treatment with hot water, followed by cooling and filtration of the wash-water, is repeated three times, the washings being added to the first filtrate. The mixed washings and filtrate are next made up to 1 liter, and aliquot parts are titrated with the decinormal sodium hydroxid, and the total acidity calculated. The number so obtained represents the volume of decinormal sodium hydroxid neutralized by the soluble acids of the butter-fat taken, plus that corresponding to the excess of the standard acid used, viz., 1 c. c. The number is, therefore, to be diminished by 5, corresponding to the excess of 1 c. c. of semi-normal acid. This corrected volume, multiplied by .0088 gives the weight of butyric acid in the amount of butter-fat saponified.

The flask containing the cake of insoluble acids and the paper through which the soluble acids were filtered are allowed to drain and dry for twelve hours, when the cake, together with as much of the acids as can be removed from the filter paper, are transferred to a weighed glass dish. The funnel and filter are then set in an Erlenmeyer flask, and the filter washed thoroughly with absolute alcohol. The flask is rinsed with the washings from the filter paper, then with pure alcohol, and these transferred to the glass dish, which is placed in the steam bath, and after the alcohol is evaporated the residue is dried for two hours in an air bath at 100° C., cooled in a desiccator, and weighed. It is heated in the air bath for two hours more, cooled and weighed. If the two weighings are decidedly different, a further heating for two hours must be made. The residue is the total insoluble acids of the sample.

Saponification equivalent (*Koettstorfer number*). This is the number of milligrams of potassium hydroxid required to saponify 1 gram of fat. The lower the molecular weight of the acid radicles present, the higher the Koettstorfer number. The following is the A. O. A. C. process :—

The reagents are the same as those employed in the de-

termination of soluble and insoluble acid, except that the standard sodium hydroxid is not needed.

Between one and two grams of the sample are weighed into a saponification flask (p. 81), 25 c. c. of the alcoholic potassium hydroxid added, the flask stoppered and heated in the steam bath until the fat is entirely saponified. The operation may be aided by occasional agitation. The alkaline solution is to be always measured by the same pipet, and it should always be allowed to drain for the same length of time (thirty seconds). Several blank experiments should be conducted at the same time.

As soon as the saponification is complete, the flasks are removed from the bath, cooled, and the contents titrated with semi-normal hydrochloric acid, using phenolphthalein as indicator. The Koettstorfer number is obtained by subtracting the number of cubic centimeters of hydrochloric acid necessary to neutralize the alkali after saponification, from the number necessary to neutralize the blank, multiplying the result by 28.06, and dividing the product by the number of grams of fat used.

It is generally considered that a sample which gives a Koettstorfer number less than 226 is not pure butter-fat, and the formula proposed for calculating the probable amount of adulterant is $x = 3.17\ (227 - n)$ in which x is the percentage of adulterant, and n the Koettstorfer number. With oleomargarin, beef dripping, tallow, and lard, usually $n = 195$ to 197.

Various physical tests have been proposed, among which the most satisfactory are the viscosity and refraction index. Many observers have noted that a relation exists between the chemical composition of a liquid and the velocity of transpiration.

Viscosity.—C. Killing (abstract in *Analyst*, 1895) uses the following apparatus for the determination of viscosity. A wide glass cylinder is closed at the bottom by a rubber

stopper, through which passes a short tube, having its top ground to receive a sort of pipet, which holds about 50 c. c., and permits of the introduction of a small thermometer into the body. The upper tube of the pipet, which passes through the top stopper of the cylinder, is closed by a stopper. The pipet has three marks, one below the body and two above, the latter being placed about 1 cm. apart. The stopper at the top of the cylinder is made in two pieces, and a second thermometer is passed through one of these. The whole apparatus is fixed in a clamp, and a beaker is placed below to receive the fat.

Originally, the standard of viscosity was taken to be the time required for a definite volume of water at 20° C. to run out, but it was found that two apparatus might give identical results for water without doing so for some fat. The times will be similar only when the body of the pipet, delivery tube, etc., are exactly of the same dimensions. For this reason each apparatus must be standardized for the mean "running out" time of butter and for margarin. Killing gives the following average figures:—

Butter,	3	min.	43.5	sec.
Oleomargarin,	4	"	19	"
Lard,	4	"	28	"
Beef-fat,	4	"	33	"

Except cocoa-fat, the viscosity of which is less than that of butter-fat, the values for vegetable fats used in oleomargarin are decidedly higher. Dr. Newman Wender (*J. A. C. S.*, 1895, p. 719) has devised a form of apparatus called a Fluidometer. "The apparatus possesses, besides its inexpensiveness, other merits, chief among which is, that by means of a simple compression bulb, the liquid can be forced back and used for repeated determinations. The apparatus consists of a V-formed capillary tube, with both limbs enlarged and divided in such a manner that one arm holds 10 c. c. and the other 2 c. c. of the liquid. According to the laws of liquids in communicating tubes, the liquid flows from the wide limb, through the capillary into the smaller limb, which is placed some-

what lower. The viscosity is calculated from the time which is required for the liquid to flow from the first division to the last upper division. There is no danger of error arising from the evaporation, or contamination with foreign substances in repeating the experiments, and, furthermore, the apparatus is easily and quickly cleaned. Since it has been demonstrated that the relation between molecular weight and viscosity is not affected by solvents, Wender uses in the fluidometer a solution of the melted fat in chloroform, in order to avoid the trouble of maintaining the fat in the melted condition. The viscosity of the solvent must be taken into account, in this case. The time of transmission of the solvent is set at 100, and the calculations for solutions are based upon this." From a large number of results he gives the following figures :—

Viscosity value for pure butter,	344.30	Time,	68.8
" " " oleomargarin,	373.20	"	77.4

Every degree of temperature above 20° C. decreases the time of efflux by 1.45 seconds. A decreasing temperature retards the efflux by an average of 1.43 seconds for each degree. The results of these investigations show that the viscosimetric examination may yield as good service in distinguishing butter from oleomargarin, as any of the physical tests.

Refractive index.—J. Skalweit has made determinations of the angle of refraction of various fats, and recommends its use in detecting butter adulteration. Abbe's refractometer was employed for the determination.

The following are some of the results given :—

Water, ...	1.333
Genuine butter, ...	1.4652
" "	1.4658
Lard, ...	1.4690
Oleomargarin, 1st quality, . .	1.4692
" 2d " .	1.4720
" 3d " .	1.4796
" oil, ...	1.4680

Butterin,	1.4712	Hanoverian manufacture.
"	1.4693	
"	1.4698	
"	1.4698	
"	1.4733	English make.

An improvement on the Abbe instrument is the Oleo-refractometer of Amagat and Jean. With this, the difference between butter and its substitutes is much greater. For description of the instrument and results with various oils, see Muter, *Analyst*, 1890, and Pearmain, *Analyst*, 1895.

Neither of the above instruments gives material aid in detecting additions of *small* percentages of foreign fat to butter.

Melting point.—Genuine butter usually shows a melting point ranging from 32.6° to 34.7° C. Lard gives figures between 42° and 43° C., and "oleo-oil" from 29° to 30° C. Artificial butter may easily be prepared of the same melting point as pure butter. The determination of melting point is, therefore, of limited value in the detection of butter adulteration.

Specific gravity.—Skalweit has noted that the greatest difference between the specific gravity of butter-fat and its adulterations is found at a temperature of 35°. The temperature usually employed is that of boiling water, and the comparison is made with water at 15.5° C. as unity.

The determination is conveniently made with the Sprengel tube, which is filled by inserting the wider end in the melted fat and exerting suction. It is then placed in boiling water in a beaker, so that the capillary ends project slightly above the surface of the water. When the fat has ceased to expand, the excess is removed from the orifice by means of filter paper, the tube withdrawn from the water, dried, allowed to cool, and weighed. The

weight of fat divided by the weight of water contained by the tube at 15.5° C. gives the specific gravity.

The Westphal balance is also recommended by Estcourt for these determinations. The melted fat is contained in a wide test-tube immersed in boiling water, care being taken to protect the balance from the steam.

The specific gravity of butter-fat determined in this way usually varies between .865 and .867, while that of oleomargarin varies between .856 and .860.

Considerable use has been made of a method based upon the detection of crystalline structure by examination with polarized light. Such condition indicates, however, merely that the sample has been previously melted. By churning oleo-oil with cream, a material is obtained which shows no crystalline structure when examined in this way.

Commercial forms of oleomargarin and butter exhibit characteristic differences on heating, which may be utilized for rapidly sorting a collection of samples. When butter is heated in a small tin dish directly over a gas flame, it melts quietly, foams, and may run over the dish. Oleomargarin, under the same conditions, sputters noisily as soon as heated and foams but little. Even mixtures of butter and other fats show this sputtering action to a considerable extent. The effect depends upon the condition in which the admixed water exists, and the test is not applicable to butter which has been melted and reworked.

An alcoholic solution of sodium hydroxid, heated for a moment with butter, and then emptied into cold water, gives a distinct odor of pineapples (due to ethyl butyrate), while oleomargarin gives only the alcoholic odor.

E. A. de Schweinitz and J. A. Emery (*J. A. C. S.*, Feb., 1896) have found that the heat of combustion of butter-

fat differs notably from that of other fats. They regard the method as capable of detecting small amounts of adulteration. It requires a special form of calorimeter which is not described in the article.

W. Arnold (quoted in *Druggist's Circular*, September, 1896) states that butter-fat is less transparent to X-rays than many common fats, but the fact has not yet been applied in a practicable form for analytic work.

Butter Colors.—Various vegetable colors have been used in butter, especially turmeric and annotto, which are still employed, but coal-tar colors are rapidly replacing them. One of the first of this class to be used was "butter-yellow" (dimethylamidoazobenzene), but latterly other azo-colors readily soluble in oils have been employed. Dairymen now use mostly proprietary preparations, but the composition of these articles is liable to change, without notice, as cheaper or more suitable colors are discovered. Those now generally in use can scarcely be regarded as dangerous to health, since apart from the fact that few coal-tar colors have appreciable toxic action, the quantity used in the butter is very small. The normal coloring matter of butter is not soluble in alcohol.

The following test, described by E. W. Martin, we have found very satisfactory. Dissolve 2 parts of carbon disulfid in 15 parts of alcohol, by adding small portions of the disulfid to the alcohol and shaking gently; 25 c. c. of this mixture are placed in a convenient tube, 5 grams of the butter-fat added, and the tube shaken. The disulfid falls to the bottom of the tube, carrying with it the fatty matter, while any artificial coloring matter remains in the alcohol. The separation takes place in from one to three minutes. If the amount of the coloring matter is small

more of the fat may be used. If the alcoholic solution be evaporated to dryness and the residue treated with concentrated sulfuric acid, annotto will be indicated by the production of a greenish-blue color. With many samples of oleomargarin a pink tint will be obtained which indicates a coal-tar color.

CHEESE.

Cheese is the curd of milk which has been separated from it, pressed, and undergone some fermentation. The precipitation is produced either by allowing the milk to become sour—when the lactic acid is the agent—or by rennet. The first named method is mainly applied to the manufacture of so-called Dutch or sour-milk cheese, green Swiss cheese, and cottage cheese. More commonly cheese is obtained by means of rennet derived from the fourth stomach of the calf. The action is due to a non-organized ferment (enzym) rennin, which acts directly on the proteids and does not produce its effect through the intervention of acids. (See p. 11.) The curd (cheese) undergoes, by keeping, various decompositions, some essentially putrefactive, and due to the action of microbes. The decomposition of the cheese is termed "ripening." In the ripening of some cheeses, moulds as well as bacteria play an important part. Thus, in the manufacture of Roquefort cheese, mouldy bread is introduced between the layers of curd, and the surrounding atmosphere is kept moist to assist in the growth and development of the fungi.

In the sour-milk cheeses, ripening is restricted intentionally, since there is liability to an irregular and miscellaneous bacterial growth by which the fermentations may be carried too far, undesirable and even harmful products being formed. Such cheeses are intended for immediate use.

Cheese contains no casein, if by this term is meant the proteid as it exists in milk, or when precipitated from milk by acids. When milk is coagulated by rennet, only a part of the casein is found in the curd; and, whereas, true casein contains about 15.7 per cent. of nitrogen, the proteid matter of cheese contains about 14.3 per cent. Under the process of ripening this is further decomposed, amido- and ammonium compounds being formed, and possibly proteoses. The following figures, obtained by Van Slyke, will serve to give some idea of the extent to which the curd is changed in ripening. The figures represent average percentage on the total nitrogen. The cheese in question was an American cheddar.

	Green Cheese.	*After five months.*
Soluble nitrogen compounds,	4.23	35.52
" amido "	None	11.66
" ammonium "	None	2.92

Van Slyke's experiments seem also to indicate that the cheese ripened more rapidly when the curd was precipitated by a larger quantity of rennet and especially, that cheese rich in fat ripened more rapidly than skim-milk cheese.

The extent and character of the decomposition of the curd varies greatly in the different varieties of cheese, the fermentative process in each case being carried on by a different group of microorganisms. In the harder cheeses the decomposition is slower and less complete than in the softer ones, in which the fermentation process is of an intensely active character. Recent progress in the study of the bacteriology of cheese indicates that at no distant date it will be possible to control the quality of the article

by inoculating the milk or curd with pure cultures of microorganisms.

In addition to the fat and nitrogenous compounds just mentioned, cheese may contain a small amount of milk-sugar and of lactic and other organic acids. There is present also a certain proportion of mineral matter, alkaline and earthy phosphates, along with any salt that has been added. Traces of nitrates have been found.

Skimmed milk is not infrequently used for the production of cheese. Foreign fats, such as are used in the manufacture of oleomargarin, are sometimes incorporated, the article being often known as "filled cheese."

The analytic points usually determined in regard to cheese, are amounts of water, fat, casein, ash, the presence of fats other than butter-fat, and coloring matters.

In addition to this, especially in comparing the qualities of genuine cheeses, the proportion of proteic, amidic, and ammoniacal nitrogen is of value.

Care should be taken to select for analysis a sample which represents the average composition of the entire cheese. A thin section, reaching to the center, is preferable, and portions from various parts of this should be cut fine and mixed. This should be done with as little exposure to air as possible, to avoid loss of water.

The following methods for water, fat, ash, and total nitrogen are provisionally adopted by the A. O. A. C.:—

Water.—From 5 to 10 grams of cheese should be taken and placed in thin slices in a weighed platinum or porcelain dish which contains a small quantity of freshly ignited asbestos, to absorb the fat which may run out of the cheese. The mass is then heated in a water oven for ten hours, and weighed; the loss in weight is to be considered as water.

Ash.—The dry residue from the water determination may be taken for the ash. If the cheese be rich, the asbestos will be saturated therewith. This mass may be ignited carefully, and the fat allowed to burn off, the asbestos acting as a wick. No extra heating should be applied during the operation, as there is danger of spurting. When the flame has died out, the burning may be completed in a muffle at low redness. When desired, the salt may be determined in the ash by titration with silver nitrate and potassium chromate.

Fat (Ether extract).—Five to 10 grams of the sample are ground in a small mortar with about twice the weight of anhydrous copper sulfate. The grinding should be continued until the cheese is finely pulverized and evenly distributed throughout the mass, which will have a uniform blue color. This mixture is transferred to a glass tube which has strong filter paper, supported by a piece of muslin, tied over the end. A little of the clean anhydrous copper sulfate is put into the tube next to the filter before introducing the mixture containing the cheese. On top of the mixture is placed a tuft of ignited asbestos, and the contents of the tube extracted with anhydrous ether in the continuous extraction apparatus, for fifteen hours. The ether is removed as usual and the fat dried at 212° F., to a constant weight. (The fat-free thimbles noted in connection with the description of the Adams process will probably be found convenient substitutes for the glass tube and filter. The fat-free residue should be tested for starch.)

Nitrogen.—The nitrogen of about two grams of the cheese is determined by the Kjeldahl-Gunning method.

The above processes may be advantageously modified in some respects. The determination of water may be made by the extraction of the cheese with alcohol and ether and drying of the alcohol-ether extract and fat-free solids separately. Blyth recommends this method as more accurate and less tedious than the direct drying. In the

determination of ash, it will be better to extract the charred mass with water and proceed as described in the determination of the ash of milk.

The fat extracted by ether may be examined for other than butter-fat by the Reichert method in the usual way. When the composition of the fat is alone desired, it may often be extracted by simpler methods. Pearmain and Moor recommend that 50 grams be chopped fine and tied up in a muslin bag, which is placed in a water-bath. When the water is heated the fat will generally run out clear. If not clear it can be filtered through paper.

O. Henzold (abstract in *Analyst*, 1895) suggests the following: Three hundred grams of the powdered cheese are agitated in a wide-neck flask with 700 c. c. of 5 per cent. solution of potassium hydroxid previously warmed to 20° C. In about ten minutes the cheese dissolves, the fat floats, and by cautious shaking may be collected in lumps. The liquid is diluted, the fat removed, washed in very cold water, kneaded as dry as possible, melted, and filtered. It is claimed that the fat is not altered in composition by the process.

The fat of cheese may be estimated by the centrifugal method, as follows:—

About 3 grams of the mixed cheese are weighed and transferred to the bottle, the last portions being washed in with the aid of water. A few drops of ammonium hydroxid are added, and sufficient water to make the liquid about 15 c. c. The liquid is warmed with occasional shaking until the cheese is well disintegrated, and then treated as a sample of milk. The percentage of fat is found by multiplying the percentage reading by 15.45 and dividing by the number of grams of cheese taken for analysis.

Chattaway, Pearmain, and Moor use the following modification: Two grams of the cheese are placed in a small dish and heated on the water-bath with 30 c. c. of concentrated hydrochloric acid until a dark, purplish-colored solution is produced. The mixture is now poured into the test-bottle, portions of solution remaining in the dish rinsed with the hydrochloric acid fusel-oil mixture into the bottle, and, finally, enough strong hot acid added to fill the bottle up to the mark. It is then whirled for about a minute.

Bondzynski (*Analyst*, abst.) applies the Werner-Schmid method to the determination of fat in cheese, as follows: A weighed quantity of the finely-shredded cheese is placed in the tube and decomposed with 20 c. c. hydrochloric acid of specific gravity 1.1, containing about 19 per cent. HCl. On cautiously warming over wire gauze, the melted fat rises to the surface. After cooling, 30 c. c. of ether are added and the tube warmed very gently until the acid and ethereal solution of fat separate sharply. Centrifugal force helps this, but is not essential. After the volume of ether has been read off, 20 c. c. are pipetted off into a weighed Erlenmeyer flask. From this the quantity of fat in the entire solution may be calculated.

Determination of proteid nitrogen (Stutzer's method).—Place 0.7 to 0.8 gram of the cheese in a beaker, heat to boiling, add two or three c. c. of saturated alum solution (to decompose alkaline phosphate), add a quantity of copper hydroxid mixture prepared as below, containing about 0.5 gram of the hydroxid, and stir thoroughly; filter when cold, wash with cold water, and without removing the precipitate from the filter determine the nitrogen by the Kjeldahl-Gunning method. Before distillation, sufficient

potassium sulfid solution must be added to precipitate the copper.

Copper hydroxid mixture.—Dissolve 20 grams of pure crystallized copper sulfate in one liter of water, and add five c. c. glycerol; add dilute solution of sodium hydroxid until the solution is alkaline; filter; rub the preparation up with water containing five c. c. of glycerol per liter, and then wash by decantation or filtration until the washings are no longer alkaline. Rub the preparation up again in a mortar with water containing ten per cent. glycerol, thus preparing a uniform gelatinous mass that may be measured with a pipette. Determine the quantity of copper hydroxid per centimeter of this mixture.

Another method is to heat a portion of the cheese with water, precipitate the soluble proteids by lead acetate, and determine separately the nitrogen in the insoluble portion and lead acetate precipitate.

Ammonium compounds.—About five grams of cheese are rubbed up in a mortar with water, transferred to a filter, and washed with a liter of cold water. The filtrate concentrated by boiling (if alkaline, it must be neutralized before heating), magnesia added, the liquid distilled, and the ammonium hydroxid in the distillate estimated by titration with standard acid.*

Amido-compounds.—The nitrogen as amido-compounds is estimated by subtracting from the figure for total nitrogen, the sum of the proteid and ammoniacal nitrogen. If nitrates are present, the nitrogen as such should also be estimated and subtracted.

Chrome yellow has been found in the rind of cheese. It may be detected by ashing the sample in a porcelain crucible, assisting the burning of the carbon by a little nitric

acid, and applying the usual tests for lead and chromium.

ANALYSES OF VARIOUS CHEESES.

(Reports by Chattaway, Pearmain, and Moor.)

Name.	Water.	Ash.	Fat.	Reichert Number.	N.
Cheddar,	33.0	4.3	29.5	24.2	4.31
Gorgonzola,	40.3	5.3	26.1	22.1	4.36
Dutch,	41.8	6.3	10.6	27.0	5.11
Gruyère,	28.2	4.7	28.6	30.0	4.93
Stilton,	19.4	2.6	42.2	29.0	4.73
Cheshire,	37.8	4.2	31.3	31.6	4.03
Gloucester,	33.1	5.0	23.5	31.4	4.99
Camembert,	47.9	4.7	41.9	31.0	3.83
Parmesan,	32.5	6.2	17.1	28.0	6.86
Roquefort,	29.6	6.7	30.3	36.8	4.45
Double Cream,	57.6	3.4	39.3	31.2	3.14
American,	30.6	3.6	27.7	3.0	4.84

The last sample is "filled" cheese.

Analyses of "Cacio-cavallo" (Horse-cheese, a very popular cheese in southern Italy), made from whole milk. (G. Sartori, *Staz. Sper. Ag. Ital.*, xxii, 337. *Analyst*, abstract, 1893):—

Water,	19.756
Fat,	36.706
Total proteids,	37.825
Ash (without NaCl),	2.340
Salt,	3.260
Total,	99.887
Pure proteids,	34.125
Ammoniacal nitrogen,	.0616
Amidic nitrogen,	.665
Reichert-Wollny figure for fat,	25.30

FERMENTED MILK PRODUCTS.

The usual fermentation of milk is the conversion of the lactose into lactic acid, but by special methods other changes may be substituted. These modified fermentations are of rather ancient origin, and being produced by mixture of organisms, the products are complex and irregular. The proteids are more or less changed into proteoses and peptones. With advances in mycology, it will be possible to carry out each fermentation by a pure culture and thus obtain a product of any desired composition.

Kumiss is milk which has undergone alcoholic fermentation. The inhabitants of the steppes of Russia prepare it from mare's milk. When cow's milk is used, cane-sugar must be added. It is often made by adding cane-sugar and yeast to skim-milk.

Vieth (*Analyst*, 1885 and 1886) gives the following analyses of kumiss:—

KUMISS FROM COW'S MILK.

At the End of	Alcohol.	Solids.	Fat.	Casein.	Albumin.	Lactoproteid and peptone.	Lactic Acid.	Sugar.	Ash.	
									Soluble	Insoluble.
One day, .	1.12	11.33	1.65	2.06	.30	.32	.26	6.16	.16	.42
One week, .	.92	8.93	1.48	2.00	.22	.56	.97	3.14	.22	.34
Three weeks,	1.03	8.66	1.58	1.93	.21	.74	1.39	2.23	.23	.35
Three months	1.12	8.52	1.57	1.70	.09	.91	1.94	1.73	.25	.33

The item "lactoproteid and peptone" refers to the sub-

stances precipitated by tannin after removal of the casein and albumin.

KUMISS FROM MARE'S MILK.

At the end of	*Alcohol*	*Fat*	*Nitrogenous Matters*	*Lactic Acid*	*Sugar*	*Ash*
1 day, .	2.47	1.08	2.25	0.64	2.21	0.36
8 days,	2.70	1.13	2.00	1.16	0.69	0.37
22 " . .	2 84	1.27	1.97	1.26	0.51	0.36

Kefyr.—This is usually made from cow's milk. It has been used in the Caucasus for centuries. For its preparation a peculiar ferment is used, which is contained in the kefyr grains. These are first soaked in water, by which they are caused to swell, and are rendered more active and then added to the milk. If taken out of the milk and dried, the grains may be used repeatedly.

The following analysis is given by Hammarsten (quoted by Aikman, "Milk, Its Nature and Composition"):—

Alcohol,	0.72
Fat,	3.08
Casein,	2.94
Albumin,	0.186
Peptones,	0.067
Lactose,	2.685
Lactic Acid,	.727
Ash,	.708

According to König, good kefyr will not contain more than one per cent. of lactic acid.

APPENDIX.

A

VIETH'S TABLE FOR CORRECTING THE SPECIFIC GRAVITY OF MILK FOR TEMPERATURE, EXTENDED BY COCHRAN.

The figures for temperatures from 38° to 45° inclusive, were determined by experiment by C. B. Cochran.

DIRECTIONS FOR USE.

Find the Temperature of the Milk in the uppermost horizontal line, and the Specific Gravity in the first vertical column. In the same line with the latter, under the Temperature, is given the Corrected Specific Gravity.

Sp. Gr.	Fahrenheit Degrees.							
	38	39	40	41	42	43	44	45
1027	25.2	25.3	25.3	25.4	25.5	25.6	25.7	25.8
28	26.1	26.2	26.2	26.3	26.4	26.5	26.6	26.7
29	27.1	27.2	27.2	27.3	27.4	27.5	27.6	27.7
30	28.0	28.1	28.1	28.2	28.3	28.4	28.5	28.6
31	29.0	29.1	29.1	29.2	29.3	29.4	29.5	29.5
32	29.9	30.0	30.0	30.1	30.1	30.2	30.3	30.4
33	30.7	30.7	30.7	30.9	31.0	31.1	31.2	31.3
34	31.6	31.7	31.7	31.9	32.0	32.1	32.2	32.2
35	32.5	32.6	32.7	32.9	32.9	33.0	33.0	33.0
36	33.5	33.6	33.7	33.8	33.9	34.0	34.0	34.0
37	34.5	34.6	34.7	34.8	34.9	35.0	35.0	35.0
38	35.4	35.5	35.6	35.7	35.8			

Sp. Gr.	Fahrenheit Degrees.									
	46	47	48	49	50	51	52	53	54	55
1020	19.0	19.1	19.1	19.2	19.2	19.3	19.4	19.4	19.5	19.6
21	20.0	20.0	20.1	20.2	20.2	20.3	20.3	20-4	20.5	20.6
22	21.0	21.0	21.1	21.2	21.2	21.3	21.3	21.4	21.5	21.6
23	22.0	22.0	22.1	22.2	22.2	22.3	22.3	22.4	22.5	22.6
24	22.9	23.0	23.1	23.2	23.2	23.3	23.3	23.4	23.5	23.6
25	23.9	24.0	24.0	24.1	24.1	24.2	24.3	24.4	24.5	24.6
26	24.9	24.9	25.0	25.1	25.1	25.2	25.2	25.3	25.4	25.5
27	25.9	25.9	26.0	26.1	26.1	26.2	26.2	26.3	26.4	26.5
28	26.8	26.8	26.9	27.0	27.0	27.1	27.2	27.3	27.4	27.5
29	27.8	27.8	27.9	28.0	28.0	28.1	28.2	28.3	28.4	28.5
30	28.7	28.7	28.8	28.9	29.0	29.1	29.1	29.2	29.3	29.4
31	29.6	29.6	29.7	29.8	29.9	30.0	30.1	30.2	30.3	30.4
32	30.5	30.5	30.6	30.7	30.9	31.0	31.1	31·2	31.3	31.4
33	31.4	31.4	31.5	31.6	31.8	31.9	32.0	32.1	32.3	32.4
34	32.3	32.3	32.4	32.5	32.7	32.9	33.0	33.1	33.2	33.3
35	33.1	33.2	33.4	33.5	33.6	33.8	33.9	34.0	34.2	34.3

Sp. Gr.	Fahrenheit Degrees.									
	56	57	58	59	60	61	62	63	64	65
1020	19.7	19.8	19.9	19.9	20.0	20.1	20.2	20.2	20.3	20.4
21	20.7	20.8	20.9	20.9	21.0	21.1	21.2	21.3	21.4	21.5
22	21.7	21.8	21.9	21.9	22.0	22.1	22.2	22.3	22.4	22.5
23	22.7	22.8	22.8	22.9	23.0	23.1	23.2	23.3	23.4	23.5
24	23.6	23.7	23.8	23.9	24.0	24.1	24.2	24.3	24.4	24.5
25	24.6	24.7	24.8	24.9	25.0	25.1	25.2	25.3	25.4	25.5
26	25.6	25.7	25.8	25.9	26.0	26.1	26.2	26.3	26.5	26.6
27	26.6	26.7	26.8	26.9	27.0	27.1	27.3	27.4	27.5	27.6
28	27.6	27.7	27.8	27.9	28.0	28.1	28.3	28.4	28.5	28.6
29	28.6	28.7	28.8	28.9	29.0	29.1	29.3	29.4	29.5	29.6
30	29.6	29.7	29.8	29.9	30.0	30.1	30.3	30.4	30.5	30.7
31	30.5	30.6	30.8	30.9	31.0	31.2	31.3	31.4	31.5	31.7
32	31.5	31.6	31.7	31.9	32.0	32.2	32.3	32.5	32.6	32.7
33	32.5	32.6	32.7	32.9	33.0	33.2	33.3	33.5	33.6	33.8
34	33.5	33.6	33.7	33.9	34.0	34.2	34.3	34.5	34.6	34.8
35	34.5	34.6	34.7	34.9	35.0	35.2	35.3	35.5	35.6	35.8

Sp. Gr.	Fahrenheit Degrees.									
	66	67	68	69	70	71	72	73	74	75
1020	20.5	20.6	20.7	20.9	21.0	21.1	21.2	21.3	21.5	21.6
21	21.6	21.7	21.8	22.0	22.1	22.2	22.3	22.4	22.5	22.6
22	22.6	22.7	22.8	23.0	23.1	23.2	23.3	23.4	23.5	23.7
23	23.6	23.7	23.8	24.0	24.1	24.2	24.3	24.4	24.6	24.7
24	24.6	24.7	24.9	25.0	25.1	25.2	25.3	25.5	25.6	25.7
25	25.6	25.7	25.9	26.0	26.1	26.2	26.4	26.5	26.6	26.8
26	26.7	26.8	27.0	27.1	27.2	27.3	27.4	27.5	27.7	27.8
27	27.7	27.8	28.0	28.1	28.2	28.3	28.4	28.6	28.7	28.9
28	28.7	28.8	29.0	29.1	29.2	29.4	29.5	29.7	29.8	29.9
29	29.8	29.9	30.1	30.2	30.3	30.4	30.5	30.7	30.9	31.0
30	30.8	30.9	31.1	31.2	31.3	31.5	31.6	31.8	31.9	32.1
31	31.8	32.0	32.2	32.2	32.4	32.5	32.6	32.8	33.0	33.1
32	32.9	33.0	33.2	33.3	33.4	33.6	33.7	33.9	34.0	34.2
33	33.9	34.0	34.2	34.3	34.5	34.6	34.7	34.9	35.1	35.2
34	34.9	35.0	35.2	35.3	35.5	35.6	35.8	36.0	36.1	36.3
35	35.9	36.1	36.2	36.4	36.5	36.7	36.8	37.0	37.2	37.3

B

TOTAL SOLIDS CALCULATED FROM FAT AND SPECIFIC GRAVITY.

Formula of Hehner and Richmond, corrected by H. Droop Richmond.

$$\text{T. S.} = \frac{G}{4} + \frac{6}{5} F + 0.14.$$

Sp. Gr.	Fat.									
	1.0	1.1	1.2	1.3	1.4	1.5	1.6	1.7	1.8	1.9
1030.5	8.96	9.08	9.20	9.32	9.44	9.56	9.68	9.80	9.92	10.04
31.0	9.09	9.21	9.33	9.45	9.57	9.68	9.80	9.92	10.04	10.16
.5	9.21	9.33	9.45	9.57	9.69	9.81	9.93	10.05	10.17	10.29
32.0	9.34	9.46	9.58	9.70	9.82	9.94	10.06	10.18	10.30	10.42
.5	9.46	9.58	9.70	9.82	9.94	10.06	10.18	10.30	10.42	10.54
33.0	9.59	9.71	9.83	9.95	10.07	10.19	10.31	10.43	10.55	10.67
.5	9.71	9.83	9.95	10.07	10.19	10.31	10.43	10.55	10.67	10.79
1034.0	9.84	9.96	10.08	10.20	10.32	10.44	10.56	10.68	10.80	10.92

Sp. Gr.	Fat									
	2.0	2.1	2.2	2.3	2.4	2.5	2.6	2.7	2.8	2.9
1026.0	9.04	9.16	9.28	9.40	9.52	9.64	9.76	9.88	10.00	10.12
.5	9.17	9.29	9.41	9.53	9.65	9.77	9.89	10.01	10.13	10.25
27.0	9.29	9.41	9.53	9.65	9.77	9.89	10.01	10.13	10.25	10.37
.5	9.42	9.54	9.64	9.76	9.88	10.00	10.12	10.24	10.36	10.48
28.0	9.54	9.66	9.78	9.90	10.02	10.14	10.26	10.38	10.50	10.62
.5	9.67	9.79	9.91	10.03	10.15	10.27	10.39	10.51	10.63	10.75
29.0	9.79	9.91	10.03	10.15	10.27	10.39	10.51	10.63	10.75	10.87
.5	9.92	10.04	10.16	10.28	10.40	10.52	10.64	10.76	10.88	11.00
30.0	10.04	10.16	10.28	10.40	10.52	10.64	10.76	10.88	11.00	11.12
.5	10.17	10.29	10.41	10.53	10.65	10.77	10.89	11.01	11.13	11.25
31.0	10.29	10.41	10.53	10.65	10.77	10.89	11.01	11.13	11.25	11.37
.5	10.42	10.54	10.66	10.78	10.90	11.02	11.14	11.26	11.38	11.50
32.0	10.54	10.66	10.78	10.90	11.02	11.14	11.26	11.38	11.50	11.62
.5	10.67	10.79	10.91	11.03	11.15	11.27	11.39	11.51	11.63	11.75
33.0	10.80	10.92	11.04	11.16	11.28	11.40	11.52	11.64	11.76	11.88
.5	10.92	11.04	11.16	11.28	11.40	11.52	11.64	11.76	11.88	12.00
1034.0	11.04	11.16	11.28	11.40	11.52	11.64	11.76	11.88	12.00	12.12

Sp. Gr.	Fat.									
	3.0	3.1	3.2	3.3	3.4	3.5	3.6	3.7	3.8	3.9
1026.0	10.24	10.36	10.48	10.60	10.72	10.84	10.96	11.08	11.20	11.32
.5	10.37	10.49	10.61	10.73	10.85	10.97	11.09	11.21	11.33	11.45
27.0	10.49	10.61	10.73	10.85	10.97	11.09	11.21	11.33	11.45	11.57
.5	10.62	10.74	10.86	10.98	11.10	11.22	11.34	11.46	11.58	11.70
28.0	10.74	10.86	10.98	11.10	11.22	11.34	11.46	11.58	11.70	11.82
.5	10.87	10.99	11.11	11.23	11.35	11.47	11.59	11.71	11.83	11.95
29.0	10.99	11.11	11.23	11.35	11.47	11.59	11.71	11.83	11.95	12.07
.5	11.11	11.23	11.35	11.47	11.59	11.71	11.83	11.95	12.07	12.19
30.0	11.24	11.36	11.48	11.60	11.72	11.84	11.96	12.08	12.20	12.32
.5	11.37	11.49	11.61	11.73	11.85	11.97	12.09	12.21	12.33	12.45
31.0	11.49	11.61	11.73	11.85	11.97	12.09	12.21	12.33	12.45	12.57
.5	11.62	11.73	11.85	11.97	12.09	12.21	12.33	12.45	12.57	12.69
32.0	11.74	11.86	11.98	12.10	12.22	12.34	12.46	12.58	12.70	12.82
.5	11.87	11.99	12.11	12.23	12.35	12.47	12.59	12.71	12.83	12.95
33.0	11.99	12.11	12.23	12.35	12.47	12.59	12.71	12.83	12.95	13.07
.5	12.12	12.24	12.36	12.48	12.60	12.72	12.84	12.96	13.08	13.20
1034.0	12.24	12.36	12.48	12.60	12.72	12.84	12.96	13.08	13.20	13.32

Sp. Gr.	Fat.									
	4.0	4.1	4.2	4.3	4.4	4.5	4.6	4.7	4.8	4.9
1026.0	11.44	11.56	11.68	11.80	11.92	12.04	12.16	12.28	12.40	12.52
.5	11.57	11.69	11.81	11.93	12.05	12.17	12.29	12.41	12.53	12.65
27.0	11.69	11.81	11.93	12.05	12.17	12.29	12.41	12.53	12.65	12.77
.5	11.82	11.94	12.06	12.18	12.30	12.42	12.54	12.66	12.78	12.90
28.0	11.94	12.06	12.18	12.30	12.42	12.54	12.66	12.78	12.90	13.02
.5	12.07	12.19	12.31	12.43	12.55	12.67	12.79	12.91	13.03	13.15
29.0	12.19	12.31	12.43	12.55	12.67	12.79	12.91	13.03	13.15	13.27
.5	12.32	12.44	12.56	12.68	12.80	12.92	13.04	13.16	13.28	13.40
30.0	12.44	12.56	12.68	12.80	12.92	13.04	13.16	13.28	13.40	13.52
.5	12.57	12.69	12.81	12.93	13.05	13.17	13.29	13.41	13.53	13.65
31.0	12.69	12.81	12.93	13.05	13.17	13.29	13.41	13.53	13.65	13.77
.5	12.82	12.94	13.06	13.18	13.30	13.42	13.54	13.66	13.78	13.90
32.0	12.94	13.06	13.18	13.30	13.42	13.54	13.66	13.78	13.90	14.02
.5	13.07	13.19	13.31	13.43	13.55	13.67	13.79	13.91	14.03	14.15
33.0	13.19	13.31	13.43	13.55	13.67	13.79	13.91	14.03	14.15	14.27
.5	13.32	13.44	13.56	13.68	13.80	13.92	14.04	14.16	14.28	14.40
1034.0	13.44	13.56	13.68	13.80	13.92	14.04	14.16	14.28	14.40	14.52

Sp. Gr.	Fat.									
	5.0	5.1	5.2	5.3	5.4	5.5	5.6	5.7	5.8	5.9
1026.0	12 64	12.76	12.88	13.00	13.12	13.24	13.36	13.48	13.60	13.72
.5	12.77	12.89	13.01	13.13	13.25	13.37	13.49	13.61	13.73	13.85
27.0	12.89	13.01	13.13	13.25	13.37	13.49	13.61	13.73	13.85	13.97
.5	13.02	13.13	13.25	13.37	13.49	13.61	13.73	13.85	13.97	14.09
28.0	13.14	13.26	13.38	13.50	13.62	13.74	13.86	13.98	14.10	14.22
.5	13.27	13.39	13.51	13.63	13.75	13.87	13.99	14.11	14.23	14.35
29.0	13.39	13.51	13.63	13.75	13.87	13.99	14.11	14.23	14.35	14.47
.5	13.52	13.64	13.76	13.88	14.00	14.12	14.24	14.36	14.48	14.60
30.0	13.64	13.76	13.88	14.00	14.12	14.24	14.36	14.48	14.60	14.72
.5	13.77	13.89	14.01	14.13	14.25	14.37	14.49	14.61	14.73	14.85
31.0	13.89	14.01	14.13	14.25	14.37	14.49	14.61	14.73	14.85	14.97
.5	14.02	14.14	14.26	14.38	14.50	14.62	14.74	14.86	14.98	15.10
32.0	14.14	14.26	14.38	14.50	14.62	14.74	14.86	14.98	15.10	15.22
.5	14.27	14.39	14.51	14.63	14.75	14.87	14.99	15.11	15.23	15.35
33.0	14.39	14.51	14.63	14.75	14.87	14.99	15.11	15.23	15.35	15.47
.5	14.51	14.63	14.75	14.87	14.99	15.11	15.23	15.35	15.47	15.59
1034.0	14.64	14.76	14.88	15.00	15.12	15.24	15.36	15.48	15.60	15.72

C

WEIN'S TABLE FOR EQUIVALENTS OF LACTOSE, CALCULATED FOR USE IN SOXHLET'S METHOD.

Copper.	Lactose.	Factor.	Copper.	Lactose.	Factor.	Copper.	Lactose.	Factor.
120	86.4	.73	215	158.2	.76	310	232.2	.81
125	90.1	.73	220	161.9	.76	315	236.1	.81
130	93.8	.74	225	165.7	.76	320	240.0	.81
135	97.6	.74	230	169.4	.76	325	243.9	.81
140	101.3	.74	235	173.1	.76	330	247.7	.82
145	105.1	.74	240	176.9	.76	335	251.6	.82
150	108.8	.74	245	180.8	.77	340	255.7	.82
155	112.6	.75	250	184.8	.77	345	259.8	.82
160	116.4	.75	255	188.7	.78	350	263.9	.82
165	120.2	.75	260	192.5	.78	355	268.0	.82
170	123.9	.75	265	196.4	.78	360	272.1	.82
175	127.8	.75	270	200.3	.79	365	276.2	.82
180	131.6	.75	275	204.3	.80	370	280.5	.85
185	135.4	.76	280	208.3	.80	375	284.8	.85
190	139.3	.76	285	212.3	.80	380	289.1	.85
195	143.1	.76	290	216.3	.80	385	293.4	.85
200	146.9	.76	295	220.3	.80	390	297.7	.85
205	150.7	.76	300	224.4	.81	395	302.0	.85
210	154.5	.76	305	228.3	.81	400	306.3	.85

The weights of copper and lactose ($C_{12}H_{22}O_{11} + H_2O$) are in milligrams. For amounts of copper intermediate between those given in the table, the quantity of lactose is determined by the factor in the third column, which represents the weight of copper corresponding to 1 milligram of lactose at that point. Thus, if 178 milligrams of copper are obtained, the calculation will be $178 - 175 = 3$; $3 \times .75 = 2.25$ additional milligrams of lactose; $175 = 127.8$. $\therefore 178 = 130.00$. The figure in the second decimal place in the product may be disregarded. The equivalent weights of copper and copper oxid are almost exactly in the ratio of 4 to 5, hence, if the weight is in terms of copper oxid, it may be converted into copper by multiplying by 0.8.

Addendum to page 29.

The mixture of amyl alcohol and hydrochloric must not be drawn into the measuring pipet by suction. It should be kept in a bottle provided with a pipet which can be filled to the mark by dipping, or the Greiner overflow-pipet may be used. Rigid accuracy in the measurement of the solution is not needed.

Addendum to page 46.

N. Leonard (*Analyst*, June, 1896) finds that the addition of a trace of ferric chlorid to the sulfuric acid increases the delicacy of Hehner's test for formaldehyde.

INDEX.

Catalogue No. 8. February, 1898.

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